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RECENT CONCEPTS IN THE TREATMENT OF HEPATIC DISEASE[1]

OSCAR D. RATNOFF

From the Department of Medicine, The Johns Hopkins University School of Medicine, Baltimore, Maryland

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During the last dozen years, the treatment of cirrhosis of the liver has undergone a radical change. This change has resulted in some improvement in the life expectancy of patients with cirrhosis, though not as much as one would like to see. This is due undoubtedly in large measure to our limited understanding of the nature of cirrhosis, but it is due also to the fact that patients present themselves for treatment only after the disease is well advanced.

During the nineteenth century, the usual treatment for cirrhosis of the liver included abstinence from the use of what were usually called spirits, and a diet consisting of fruits, leguminous vegetables, cereals, milk and eggs. The patient was cupped and fed laxatives and diuretics. Finally, as a last resort, abdominal paracentesis was performed (1). The prognosis was almost uniformly bad. Early in the twentieth century, experimental evidence began to appear which seemed to indicate that the ingestion of meat and of fat was harmful to patients with liver disease, whereas carbohydrates exerted a beneficial effect. This view was based on the following types of experimental data. The portal vein of dogs was cut off from the liver and its blood diverted to the vena cava; that is, an Eck fistula was formed. Then, when these dogs were fed raw meat they became severely and often fatally ill (2). Opie (3) demonstrated that when rats were injected with phosphorus they usually survived if fed a high carbohydrate diet composed of oats and sugar, whereas they usually died if fed a diet limited to meat, and would occasionally die if fed a diet limited to fat. When the rats were injected with chloroform instead of phosphorus, a fatty diet seemed to be most injurious. Later, Mann and Magath (4) reported that the administration of glucose delayed the

[1] Presented at the Saturday morning Therapeutic Conference, February 7, 1948.

death of dogs whose livers had been removed. Again, Bollman and Mann (5) showed that dogs injected with carbon tetrachloride would die of cirrhosis if fed a diet limited to meat, but would survive the experimental period if fed milk, bread and syrup. All these experiments, and many more like them, led to the treatment of both acute and chronic liver disease with diets rich in carbohydrates and low in proteins and fats. Fig. 1 demonstrates the prognosis of 296 patients with cirrhosis who had ascites, at four hospitals under supposedly ex-

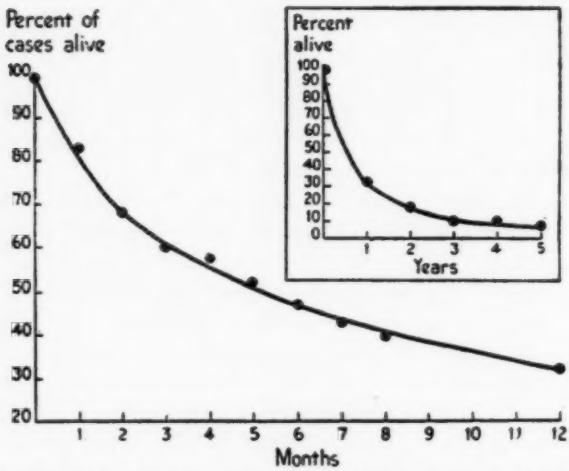

FIG. 1. Survival after the onset of ascites in 296 patients with Laennec's cirrhosis treated between 1916 and 1938 (6).

cellent conditions, between 1916 and 1938 (6). Most of these patients were treated with a diet rich in carbohydrate and poor in protein and fat.

In 1937, Patek (7) reported results with a radical departure in therapy. He was impressed by the view, fresh at that time, that the infirmities of alcoholics such as polyneuritis, pellagra, and beriberi were due not to the alcohol consumed but to the concomitant poor dietetic intake of these patients. At the time, there were many enthusiastic reports in the literature concerning dramatic cures of alcoholic polyneuritis with B vitamins. Patek undertook, therefore, to treat patients with

cirrhosis with a diet rich in B vitamins, supplemented by oral and parenteral B vitamins. Ten of thirteen patients fed a balanced diet of approximately 300 grams of carbohydrate, 100 grams of protein, and 120 grams of fat showed striking improvement. This seemed well beyond the chance expectation of improvement in this disease. Patek appreciated the fact that the value of the diet given these patients did not necessarily depend upon any single component, and in further studies an even higher caloric intake of protein, carbohydrate and fat was given. Thus the dietetic regimen of patients under Patek's care now includes approximately 350 grams of carbohydrates, 140 grams of protein, and 175 grams of fat, a total of 3500 calories a day (8). The diet consists chiefly of meat, fish or poultry, milk, eggs, fruit and green vegetables. The diet is supplemented with a total of 50 grams of powdered brewer's yeast a day, or, if this is not tolerated, an oral B complex preparation. In addition, intramuscular injection of thiamine chloride, 5 mgm. daily, and unconcentrated liver extract, 5 cc. twice weekly, is given. The diets are diversified according to the patient's individual desires, rather than to fit the general hospital routine, since food that is not eaten is of no value to the patient. Furthermore, the patient is spoon fed when necessary by the attendants, nurses and residents, in order to coax the food down the esophagus. Fig. 2 shows a recent tabulation of the prognosis of patients with cirrhosis and ascites who were treated with this regime. The survival of these patients is compared with 230 patients of the 1916–1938 series who were alive one month after the onset of ascites. It should be pointed out that the improvement in the prognosis of patients with cirrhosis occurred during the era of chemotherapy for infection, and this may account for some of the improvement noted.

As might be expected, the earlier a patient with cirrhosis is treated and the milder the case, the more readily improvement appears. Indeed, Post and Patek (9) were able to correlate the prognosis of patients treated with a highly nutritious diet with the plasma serum albumin level at the time of the patient's admission to the hospital. On the average, those patients who died had a serum albumin of 2.4 grams per 100 cc. on admission, whereas those who improved and whose ascites disappeared had a serum albumin averaging 3.0 grams on admission, a difference which was statistically significant. It cannot be

emphasized too strongly that the treatment of patients with cirrhosis must be continued for a long time, and the fact that improvement does not occur within several weeks should not be cause for discouragement. Fig. 3 demonstrates the length of time from the initiation of treatment until the disappearance of certain specific signs. Patek's observations have been amply confirmed by other observers (10, 11, 12).

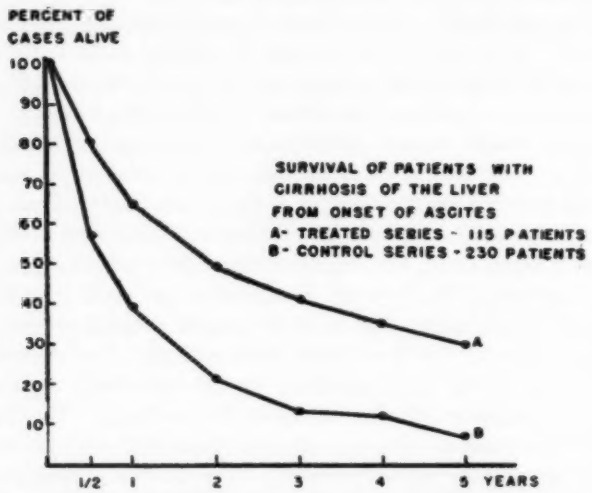

FIG. 2. Survival after the onset of ascites in 115 patients with cirrhosis of the liver treated with a highly nutritious regime, compared with 230 patients who lived at least one month after the onset of ascites, and were treated during 1916–1938 (8).

In recent years, experimental evidence has appeared which seems to place the use of a highly nutritious regime on a rational basis. In the first place, Davis and Whipple (13) had shown as early as 1919 that a diet of skim milk or casein protected dogs given chloroform, an observation confirmed in rats by Moise and Smith in 1924 (14). Then, a great flood of reports appeared, by György and Goldblatt (15), Rich and Hamilton (16), Chaikoff and Connor (17), Blumberg (18), Spellberg and Keeton (19), Webster (20) and many others, that rats, rabbits, and mice fed special diets developed necrosis of liver cells, fatty in-

filtration of the liver, and an increase in fibrous tissue in the liver. With our current hindsight, we now recognize that these diets were, in general, relatively low in protein and usually relatively high in fat as well. It was quickly found that the lesions could be prevented by increasing the protein content of the diet, or by feeding large amounts

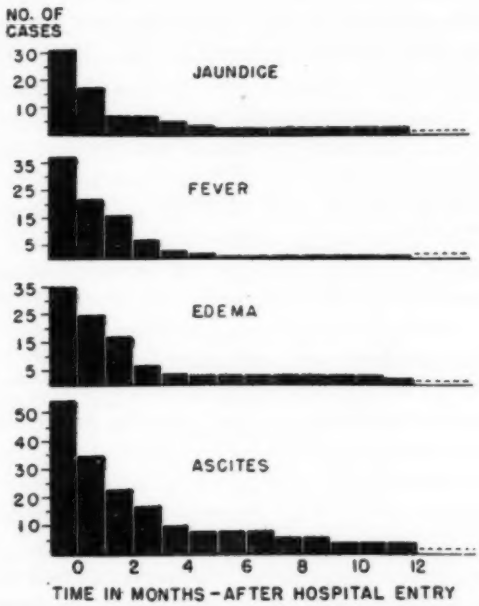

FIG. 3. The time at which specific signs of hepatic failure disappeared after the initiation of treatment with a highly nutritious regime (8).

of brewer's yeast (15, 16, 20, 21). Similarly, these substances offered more or less protection against such toxins as arsenic (22, 23) and chloroform (24).

The factors in protein and brewer's yeast which afford protection against dietetic hepatic injury are not yet clearly recognized. One can readily understand this, since the basic diet, the species of animal, and the dose of the adjuvants tested varied from study to study. For example, Daft, Sebrell and Lillie (25) reported that choline prevented

cirrhosis in rats fed a low casein, low fat, high carbohydrate diet. On the other hand, György and Goldblatt (26) found that choline would not prevent liver injury in rats fed a low casein diet when these rats got most of their calories from lard or Crisco instead of from carbohydrate. However, these rats were protected by a combination of cystine and choline. The protective action of cystine was the more remarkable since previously it had been shown that large amounts of cystine were severely toxic for the liver (27, 28). Methionine was also

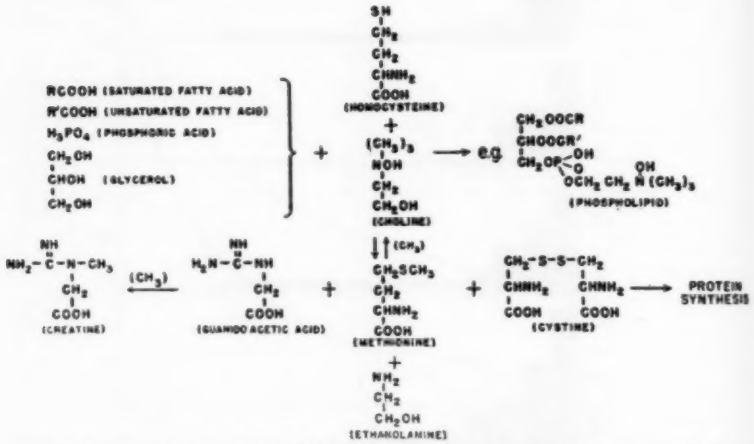

FIG. 4. Some inter-relationships between lipid transport and choline-methionine metabolism. Fat may be transported from the liver to peripheral depots as phospholipids. The choline necessary for this, however, may be needed to furnish methionine for creatine synthesis or protein synthesis. In this event, phospholipid cannot be synthesized and neutral fats accumulate in the liver.

shown to protect the liver of rats fed low casein diets (29, 30, 31), a protection potentiated by small amounts of cystine (30). It is of interest that some of the factors which seem to prevent experimental dietetic cirrhosis are the same as those which Best, Hershey (32) and others (33) found would prevent certain of the experimental fatty livers. Fig. 4 is a rough outline demonstrating some of the relationships between choline, methionine, and fat transport. To oversimplify it, fatty infiltration of the liver occurs when the neutral fats

cannot be removed from the liver by such transport systems as the phospholipids. The liver is infiltrated with fat whenever the synthesis of phospholipids is impaired. This occurs when there is an insufficiency of essential fatty acids, when there is an insufficiency of choline and methionine, when excesses of cystine cause the utilization of methionine for the formation of protein, and so forth.

The ability of lipotropic substances to prevent experimental dietetic cirrhosis was not entirely unexpected. As Connor (34) emphasized, fatty infiltration of the liver often precedes the development of cirrhosis both in human beings and experimental animals. However, it was not long before it was found that lipotropic substances did not afford complete protection against hepatic injury. For example, Daft and his associates (30) observed that rats fed a low casein diet developed hepatic necrosis as well as periportal fibrosis. Choline or methionine protected the rats against the fibrotic lesion, but had no effect on the necrotic lesion. These necrotic lesions, however, could be prevented by a combination of methionine and cystine. Himsworth and Glynn (35) also emphasized that two separate types of lesions—necrotic and fibrotic—were encountered in rats fed experimental diets. They observed that rats fed a low protein diet developed massive necrosis of an entire liver lobe. This massive necrosis occurred whether or not the liver was infiltrated with fat. Choline did not prevent the necrosis, while methionine in small amounts was completely protective (36). Now, when rats were fed a diet rich in fat and low in lipotropic substances they developed diffuse hepatic fibrosis. Here again, the pathologic sequence of fatty infiltration of the liver and fibrosis of the liver described by Connor was observed. This diffuse fibrosis could be prevented by choline (35), unlike massive hepatic necrosis. Himsworth and Glynn's experiments are not entirely free from criticism. For example, the rats were fed in groups rather than individually, so that the actual dietetic intake of each rat was not known. They were able to repeat their experiments with rats from two sources housed in two separate laboratories, but this does not entirely rule out the possibility that some indigenous infection of rats was uncovered by the low protein diets. To my knowledge, Himsworth and Glynn's experiments have not been confirmed. However, it is probably true that microscopic (rather than massive) necrosis of the liver is a lesion separate

in its origin from fibrosis. Recently Green and Brunschwig (37) demonstrated that a large series of compounds would protect rats against one or the other of the two lesions, but not necessarily both. In summary, then, hepatic lesions can be produced in experimental animals by feeding various diets, and these lesions can often be prevented by special dietetic factors. But neither the pathogenesis, prevention nor treatment of these lesions has really been clarified.

The evidence of the experimental value of choline, methionine, and combinations of choline and methionine with cystine has led to the wide use of these substances as adjuvants to a highly nutritious diet in the treatment of cirrhosis. Suffice it to say, their value under these conditions is unproved. Perhaps this is because any diversified nutritious diet is rich in the foods necessary for the nutrition of the liver. For instance, casein and egg white are said to be rich in methionine (31); wheat gliadin (38), peanut meal, muscle meats and lactalbumin in cystine (31); and meat, beef liver and pancreas, cereal, wheat flour, polished rice and rice flour, skim milk, brewer's and baker's yeast, peanut meal, soy meal and vegetables in choline (39, 40). Since these substances are present in a well balanced diet in amounts which are apparently adequate, it would seem unreasonable to expect much benefit from giving still more to the patient. It is, however, probably fair to say that choline and methionine in moderate doses are not harmful, unlike cystine which is potentially dangerous.

It should be emphasized that experimental diets deficient in these protective substances are achieved only with the greatest of care. Moreover, comparatively slight shifts in the diets of these experimental animals will cause major changes in the incidence of hepatic lesions. For example, Earle and Victor (41) showed that rats fed a diet rich in butter were more susceptible to cystine than rats fed a balanced diet, while a diet rich in lard and cod liver oil was actually slightly protective. In another situation, on the other hand, György and Goldblatt (26) observed that choline prevented hepatic necrosis in rats fed butter, but not in rats fed lard or Crisco. In another instance of the importance of the details of experimental diets, Webster (20) observed that small amounts of molasses prevented dietetic cirrhosis in rats. Molasses is a crude carbohydrate rich in inorganic sulfur and containing about 2.5 to 3 per cent protein. In the clinic, the diet is always diversified, and

the addition of one component or another does not seem to matter. Not to labor the point, but the experiments which demonstrated the toxicity of raw meat illustrate the complexity of the experimental problem very well. It was pointed out above that diets limited exclusively to lean meat aggravated the hepatic lesions produced by phosphorus (3) or carbon tetrachloride (5). Bollman and Mann (5) observed that it was not the protein of the meat which was toxic, but rather something in a water extract of the meat. More recently, Ralli and Rubin (42) reported that feeding raw lean beef to depancreatized dogs caused fatty livers, and again noted that the toxic substance was in a water extract of the meat. They discovered this extract contained inositol. When inositol was fed along with meat from which the toxic water-soluble substances had been removed, the dogs developed fatty livers. The presence of inositol, then, seems to account for some of the toxic effects of all-meat regimes. It has been shown that biotin and cholesterol produce fatty livers in rats. Just to complicate the picture further, inositol actually prevents these fatty livers produced by biotin (43) or cholesterol (44). Of course, no one advocates feeding patients a diet limited to lean meat. In the same way, arguments for the use of low fat diets are based upon experiments in which the composition of the diet is completely artificial. In practice, if the amount of fat in the diet is limited, the diet is usually so unpalatable that the patient fails to eat enough of anything.

Recently the use of intravenous liver extracts has been recommended for patients with Laennec's cirrhosis (45, 46). Liver extracts were advocated for the treatment of liver disease as early as 1626 (47). Numerous enthusiastic reports of their use appeared in the French literature early in the present century (49, 50). Recent experience from the Rockefeller Institute and elsewhere with the use of intravenous liver extract has been most encouraging. However, this study may be taken as an instance of the difficulty of interpretation of therapeutic results. In measuring the value of intravenous liver therapy, Labby and his associates (45) eliminated from consideration those patients who did not have at least a six months' course of therapy. Of those patients who had this minimal course, 77 per cent survived for two years, compared with 45 per cent who survived when treated with Patek's regime at that time (51). Now, if the data are studied, two

things become apparent. In the first place, Labby and his associates included all patients with cirrhosis in their statistics, while Patek included only those who had ascites—that is, patients in whom the disease was more advanced. In the second place, Labby and his associates eliminated from consideration those patients who did not survive six months. In other words, Labby's and Patek's data are not comparable. None of this is intended as criticism of one or the other group, but merely to emphasize the difficulties involved in making such a comparison. The use of intravenous liver extract certainly warrants further study. Patek (8) recently treated 6 patients with intravenous liver extract. These patients were in a stable state of decompensation, not responding to at least six months of the basal dietetic regime. The course of three of the patients seemed unaffected by the liver extract. The other three, who had had 33, 75, and 126 abdominal taps respectively, had an increase in appetite and their ascites gradually disappeared. Of course, this is not proof of the efficacy of liver extract. The only way that the value of specific dietetic adjuvants could be tested adequately would be to set up rigidly controlled, long term studies of such enormous proportions as to be economically unfeasible.

Thus far only the dietetic care of the patient with cirrhosis of the liver has been discussed. I should like to touch briefly on the treatment of certain specific symptoms. The factors which result in the formation of ascites are only poorly understood. For some time it was thought that the increase in portal pressure (52, 53, 54, 55) and decrease in serum albumin (56, 57, 58) and colloid osmotic pressure (59, 60) were enought to account for the presence of ascites. Ralli and her associates (61), however, pointed out that diuresis seemed to occur in some patients with cirrhosis before any rise in serum albumin or colloid osmotic pressure. They demonstrated that the urine of cirrhosis patients with ascites contained a large amount of an antidiuretic substance and suggested that this antidiuretic may contribute to ascites formation. However, this work has not been adequately confirmed. Recent work of Patek and his associates (62) suggests that differences in permeability of the portal vascular bed may determine whether or not ascites accumulates. In the treatment of ascites, an attempt is made to raise the colloid osmotic pressure of the plasma by feeding a

diet rich in protein. This is a difficult process, however. Myers and Keefer (63) and Post and Patek (64) studied the nitrogen balance of patients with cirrhosis. Although the patients retained large amounts of ingested nitrogen, the serum albumin level did not rise concomitantly. Diuretics have been used in the treatment of ascites for more than a century and are still important weapons. At present, mercurial diuretics and ammonium chloride are most frequently employed. In recent years, the effect of intravenous injection of concentrated human albumin has received extensive trial. If approximately 50 grams a day are given to patients with cirrhosis and ascites, the serum albumin level rapidly rises to normal levels (62, 65, 66), but diuresis is unimpressive. Apparently, this is because the albumin diffuses into the ascitic fluid almost as rapidly as it is injected. If larger doses of albumin are given, for example, injections of 100 grams at a time, a satisfactory diuresis is said to occur (67). Perhaps this is because the increase in plasma volume which results acts as a diuretic (60). Obviously this increase in plasma volume might be dangerous for patients with esophageal varices. Paracentesis, once reserved for the moribund, should be resorted to relatively early in the treatment of ascites, since the ascites interferes with the patient's ability to eat. It is well to remember, however, that occasionally a patient will develop cholemia after paracentesis. Although many surgical procedures have been devised for the relief of ascites, none has proved uniformly satisfactory.

Bleeding into the gastro-intestinal tract must, of course, be treated as a grave medical emergency. The patient should be put at rest, but it is wise to be cautious in the use of sedatives, since patients with impaired liver function may occasionally be more sensitive than the average person to opiates and barbiturates. Food should be withheld until signs of active bleeding have disappeared. Patients should be transfused early with whole blood, before the blood pressure drops and shock becomes irreversible. Numerous operative procedures have been suggested for the relief of portal hypertension in patients with recurrent hematemesis. For example, Blakemore (55) anastomoses the portal and systemic venous circulations—that is, he creates an Eck fistula. Eleven of nineteen patients with portal hypertension who survived the establishment of a portocaval anastomosis had no gastro-intestinal bleeding during the subsequent six months. The mortality rate of the

operation is high, being nineteen per cent in the latest analysis (68), and it is particularly high in patients whose portal hypertension is due to cirrhosis. There is, moreover, no evidence that hepatic function is improved by the operation. It is, therefore, doubtful whether this procedure will prove of value in cirrhosis except in the occasional patient whose major difficulty is recurrent hematemesis.

Cholemia, or severe hepatic decompensation, is difficult to treat. Here, perhaps, the use of glucose and vitamins parenterally is justified, for Snell and Butt (69) were able to revive six patients in hepatic coma by this means.

Hoagland and his associates (70, 71) and others (72, 73) have demonstrated that therapy has little or no effect on the course of acute infectious hepatitis. Although the classic American diet for this disease is rich in carbohydrate and poor in fat and protein, they demonstrated that a high fat, high protein regime was not only as effective, but may indeed have shortened the course of acute infectious hepatitis. The use of such adjuvants as crude liver extract and methionine was without benefit. Rest has been advocated (71, 74), and with this we agree, but the evidence which has been presented to substantiate this point of view is probably incomplete.

The question often arises whether the patient in coma from acute infectious hepatitis or cirrhosis should be given intravenous amino acids. On theoretical grounds, this might be considered dangerous since the sick liver might not be able to metabolize the excess load of amino acids. Actually, however, no deleterious effects have been observed from the intravenous use of commercially prepared amino acids in patients in hepatic coma (75) or ill with severe hepatitis or cirrhosis (77, 78), although at the same time one would hesitate to ascribe any benefit to them.

It is probably obvious that any mention of the avoidance of alcohol by patients with liver disease has been neglected until now. This is because there is no evidence that the moderate use of alcohol is deleterious if the patient at the same time eats an adequate diet (51). However, in the patient with cirrhosis, moderation is a rare virtue, and patients should be urged to become teetotallers.

Finally, it is interesting to learn that we have now, so to speak, completed the cycle. After the long period of experimenting with this or

that dietetic restriction, we have returned to the point where Murchison (79) was in 1868. The diet, he said, should include milk, eggs, farinaceous substances, plainly cooked fish and meat, that is, in the light of modern chemical analysis, a well balanced assortment of high protein foods.

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HEREDITARY MYOCLONUS EPILEPSY

Two Cases with Pathological Findings

WILLIAM I. MORSE, 2ND

From the Department of Pathology, The Johns Hopkins University and Hospital

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The literature contains only a small number of pathological reports dealing with hereditary myoclonus epilepsy. The findings lack uniformity and the pathological basis for the condition needs clarification. This report, concerning a sister and brother, presents some interesting features.

Case I. M. T.[1] (Ped. A-51076, Path. 20738) is a 7 year old white female admitted to the Harriet Lane Home on March 21, 1947, in status epilepticus.

The *family history* is of interest in that a maternal uncle died in the third decade in status epilepticus, having had grand mal seizures throughout his life. A brother G. T. constitutes the second case of this report. Three older half siblings, ages 16, 14 and 11 years are well and have never had any type of seizures.

The *past history* reveals that the child was born after a 16 hours labour requiring version and extraction. Development was rather slow. She sat at 7 months, walked at 2 years and talked at 2½ to 3 years. She has always been dull and slow. A clumsiness in walking was attributed to knock knees and pigeon feet. Measles and shingles were contracted several years before the present illness.

The *present illness* dates from April, 1946 when the child began to have head nodding attacks associated with uncontrollable jerky movements of the extremities. Episodes of falling and dropping things from her hand began 2 months later, these episodes being associated with a rolling upward of her eyes and momentary unconsciousness. On examination in the Harriet Lane Home in October, 1946 she appeared dull. An intention tremor and an unstable gait were recorded. Increased deep reflexes and a doubtful positive Babinski were noted on the right side. An electroencephalogram was interpreted as showing a petit mal and psychomotor pattern without localization. The child was improved temporarily with tridione and phenobarbital therapy. On March 21, 1947 she collapsed while at school, lost consciousness and began to have left sided convulsions which continued until admission 8 hours later.

Physical examination revealed the child to be comatose and actively convulsing. There were regular clonic contractions of the left arm and leg, increased deep

[1] The clinical aspects of these two cases will form part of a more extensive report to be published by Dr. L. Kajdi and Dr. S. Livingston.

tendon reflexes on the left, deviation of the head to the right and bilateral ankle clonus.

Pertinent *laboratory data* included a spinal fluid under increased pressure showing 15 mononuclear cells, protein of 53 mgs. per cent, a negative Wassermann and Mastic curve. The blood calcium was normal. There was no evidence by x-ray or blood analysis of lead intoxication. Ventricular air injection revealed no significant abnormality. Electroencephalogram showed a more abnormal rhythm than previously.

The convulsions were controlled with ether and paraldehyde and later the child regained consciousness, but rhythmic myoclonic jerks persisted. The jerks were first on the left side associated with a paralysis of this side which cleared slowly over a period of a month. Then the right side manifested myoclonic jerks and during the stay in hospital all the skeletal muscles were involved at some time as well as both domes of the diaphragm. These movements ceased during sleep. Five days after admission a right craniotomy was performed and revealed distended veins over the leg area and some cortical atrophy. The wound was closed without further interference. The child improved during the first month to a point where she could walk, but was not well oriented. This was followed by another episode of grand mal seizures after which she remained unconscious until her death on August 14, 1947—146 days after admission. Myoclonic jerks continued throughout this interval and there was a terminal episode of pneumonia. Numerous medications were utilized, none of which gave outstanding benefit.

Autopsy: It was performed 8 hours after death. The body weighs 20 kgs. and is 117 cms. long. The development and nutrition are normal. Aside from many small yellow foci of pneumonic consolidation in the lungs and a small mucosal nodule in the rectum, macroscopic abnormalities are confined to the head and brain.

The skull and scalp show evidence of a previous left craniotomy with satisfactory healing. The brain was injected with 4.5% formalin through the carotid arteries before removal and was hardened in 10% formalin for 40 days before sectioning. The leptomeninges are slightly thickened over the frontal and parietal lobes. The vessels of the circle of Willis are smooth and patent. There is no gross asymmetry of the brain and no gross abnormality of its fissural pattern. There is slight convolutional atrophy, more marked over the left fronto-parietal area than elsewhere. A coronal section just anterior to the foramena of Monro shows good differentiation of gray and white matter. The white matter is of normal firm texture and the fibre systems, such as the anterior limbs of the internal capsules stand out clearly. The gray matter of the basal ganglia of both hemispheres is of normal appearance and texture. The cortical gray matter of the right cerebral hemisphere averages 2½ mm. in thickness at the crown of each gyrus. On the left, from the level of the superior frontal convolution to the outer limits of the cortex of the island of Reil, the cortical gray matter is thin, measuring 1½ mm. at the crown of the gyri, and it seems to be separated from the underlying white matter by a spongy cleavage line. The ventricular system is not significantly displaced. The left ventricle

is slightly larger than the right. The ventricular linings are smooth throughout and the choroidal plexuses appear quite normal. From this plane forward to the frontal poles the left sided cortical thinning and partial detachment gradually disappears, but some signs of it can be found at almost the extreme tip of the frontal lobe. There is no other gross finding of note in the frontal lobes.

A coronal section through the anterior portion of the mammillary bodies again shows good differentiation of gray and white matter; slight enlargement of the left lateral ventricle; no enlargement of the third ventricle; and on the left side extensive cortical changes of the type described above, extending from the supero-mesial margin of the hemisphere downward to and including the left superior temporal convolution. The uncus, pyriform areas and fornices appear normal. There are two minimal areas of cortical softening on the right, but nothing to approach the extensive change on the left side. As one proceeds caudally this same phenomenon of marked cortical involvement on the left and minimal patchy involvement on the right persists to the very tips of the occipital lobes (Fig.1). The striate cortex on both sides is well preserved and a clear cut line of Gennari can be seen throughout its extent. There is, in addition, an area of softening which involves almost the entire left thalamus, apparently sparing the anterior dorsal thalamic nucleus and extending downward until it reaches the region of the corpus subthalamicum and the cerebral peduncle. Across this zone of encephalomalacia the mammo-thalamic bundle stands out sharply and is well preserved. The lentiform nuclei of both sides are grossly normal, as are the external and internal capsules and the optic tracts.

There is moderate, but definite asymmetry in the cerebellum, the right cerebellar hemisphere being throughout smaller than the left. The markings of the cerebellar folia of the right hemisphere are accentuated, and the folia themselves appear thinner than do those of the left. The central cerebellar nuclei are possibly less clearly differentiated from the central white matter than is usual, but no gross softening is palpable. The brain stem, floor of the fourth ventricle and upper cervical cord are grossly normal.

Microscopic findings: The lungs show lobular pneumonia. The liver shows fresh central necrosis and central fatty infiltration. A small benign argenaffinoma is found in the rectum. The retina and choroid of the eyes are not obtained. The remaining microscopic abnormalities are confined to the brain.

The leptomeninges are normal. The sections of cerebral cortex show foci of destruction of gray matter apparently sparing other areas completely. These lesions are more extensive on the left, and are seen in sections from all lobes on this side.

The most extreme lesion is one of such marked encephalomalacia of the cortical gray matter that even the glial framework is rudimentary except in the subpial layer (Fig. 2). There is astrocytic hyperplasia of the molecular layer, and a very imperfect meshwork of astrocytes persists in the deep layers. Compound granular corpuscles are abundant. No ganglion cells remain in such an area. The underlying white matter shows great loss of nerve fibers in a Bodian stain, and a marked

astrocytic hyperplasia. Nissl's plump astrocytes are abundant, and giant astrocytes with multiple nuclei (Fig. 3) are seen. These elements are most concentrated at the junction between gray and white matter. Fat stains reveal an abundance of fat droplets both free in the tissues and within macrophages.

Other areas show less extreme lesions, usually sparing some layers more than others. The subpial layer is always partially intact. At least a glial framework usually persists in layer II and the upper part of layer III. The ganglion cell loss is sometimes laminar in distribution, but is not consistently selective to certain layers. Frequently a few ganglion cells persist although the destructive process about them is marked. Some areas show considerable loss of neurons and marked alterations of those remaining, but the glial framework is intact and perhaps more dense than normal (Fig. 4). Such areas show some capillary proliferation. At the junction of gray and white matter a noticeable concentration of Nissl's plump astrocytes is also noted in these less severe lesions. Fat laden phagocytic cells are abundant in both gray and white matter. An early cortical change is the presence of fat laden cells in areas where the ganglion cells are normal in number and distribution. Only in some minimally affected areas are the fat laden cells obviously perivascular. All grades of ganglion cell alterations are seen up to complete cytolysis. In Nissl preparations some ganglion cells in and adjacent to the lesions show pale staining qualities, chromatolysis, cytoplasmic vacuoles, shrinkage and pyknosis of the nucleus, displacement of the nucleus and karyolysis. Some of these ganglion cell changes probably represent post mortem alterations. Numerous areas of cortex are apparently free from any lesion. Both motor cortical areas are partially destroyed. The cornua ammonis on both sides shows occasional laminar spongy areas adjacent to the ganglion cell layers. These areas differ from the above described lesions in that they show no evidence of gliosis or cellular reaction, and may represent artefact.

Myelin sheath stains show considerable loss of myelin in the white matter underlying involved areas of cerebral cortex with puddling and globule formation (Fig. 5).

The left optic thalamus shows the area of encephalomalacia noted grossly (Fig. 6) to be greatly rarified by the presence of innumerable contiguous vacuoles, so that it has become a status spongiosus (Fig. 7). Besides glial fibres the walls of this meshwork also contain nerve fibres as seen in reduced silver preparations. There is a definite increase in mature astrocytes, but no plump astrocytes are seen. The appearance suggests a more quiescent lesion of longer standing. However myelin sheath stains show puddling and globule formation. Fat stains show rather abundant fat laden phagocytes. A few foci of surviving neurons are present here and there. Some of them show chromatolysis and shrinkage. The medial portion of the right thalamus is spared, but the remainder shows the same lesion as the left. The region of the corpus subthalamicum shows essentially the same lesion as the thalamus on both sides.

The head of the left caudate nucleus shows a few small circumscribed lesions of the same type as those in the thalamus, but its greater portion is normal. The

left putamen also contains a few tiny spongy foci and a small associated area with ganglion cell loss and replacement by mature astrocytes. The pallidus on the left is normal. The section of caudate and lentiform nucleus on the right is normal.

There are circumscribed vacuolated areas in the substantia nigra on both sides, similar to those in the thalamus. The ganglion cells remain around these areas. There are lipoid laden phagocytic cells in this region, and dorsally in the decussation of the brachium conjunctivum. The pontine nuclei show very small areas of involvement by this same spongy lesion.

The cerebellar cortex is severely damaged. The more posterior portions of the hemispheres are relatively normal, but moving anteriorly toward the cerebellopontine angle, there is marked destruction and atrophy on both sides. The molecular layer becomes narrowed, the Purkinje cells disappear completely and there is marked cell loss in the granular layer of this portion. There is proliferation of the Bergmann layer. Elsewhere many of the remaining Purkinje cells show shrinkage, a coarsely granular cytoplasm, pyknosis or karyolysis. Fat stains show numerous lipoid filled macrophages in the affected gray matter, but very few in the white matter. Some of these are rod cells. Myelin sheath stains, however, show areas of puddling and globule formation in the white matter.

The dentate nucleus also shows a severe lesion (Fig. 8). The ganglion cells are somewhat decreased and there is a definite increase in mature astrocytes. The outstanding point is the presence of very numerous large vacuoles. The latter are chiefly localized at the junction between gray and white matter on both external and internal aspects of the nucleus. Many of the remaining ganglion cells show shrinkage, and some show pyknosis, displacement of the nucleus or karyolysis. Some of the cells show partial cytolysis. A fat stain reveals almost no fat laden cells in the region of the nucleus. However, a myelin sheath preparation shows areas of puddling and globule formation within and about the arc formed by the dentate nucleus.

Finally a circumscribed oval lesion 5 mm. by 2 mm. in diameter is present in the medullary substance of the cerebellum on the left side, about 4 mm. lateral to the dentate nucleus. This area is spongy and vacuolated with evidence of gliosis, and resembles the lesions described in the thalamus.

The medulla including the inferior olives is normal. The upper end of the cervical spinal cord is normal, without evidence of degeneration in the projection fibres in myelin sheath stains.

A careful search fails to reveal intracellular or extracellular inclusion bodies. Special stains for iron, including the pallidus, give a negative result. The reliability of glycogen stains is impaired by the form of fixation. Using the periodic acid routine (1), controlled by the saliva test (2), no glycogen can be demonstrated.

Case II. G. T. (Ped A-24278, Path. 19848) is an 8 year old white, male admitted to the Harriet Lane Home on Dec. 22, 1945 in status epilepticus.

The *family history* is described in the case report of sister M. T. above.

The *past history* revealed a difficult delivery requiring version and extraction,

and artificial respiration was administered for 30 minutes. Development was slow. He sat at 7 months; walked at 2 years but was clumsy. He made sentences at 3 years. The child had whooping cough at one year of age, chicken pox and measles during early childhood.

The *present illness* probably began at the age of 2½ years (20 months after the episode of whooping cough) with occasional falling spells. This was followed soon by attacks of head nodding and rhythmic movements of the arms. There were no myoclonic jerks at this time. The child was seen in the Harriet Lane Home at 4 years of age with vomiting episodes unrelated to meals or obvious psychic causes, but followed by about 2 hours of sleep. A torticollis was noted, but no other abnormalities. The vomiting improved temporarily and he was next seen at the age of 8 years. On Dec. 19, 1945, 3 days before admission there was a recurrence of vomiting and episodes of momentary unconsciousness, the eyes deviating to the right. Twelve hours later clonic jerks, chiefly right sided, began. The child became unresponsive and the jerks continued until admission.

Examination revealed the child to be comatose with occasional right sided twitches. The reflexes were depressed. After regaining consciousness examination revealed right sided weakness, intention tremor, poorly performed coordination tests and some mental retardation.

Pertinent *laboratory data* included a spinal fluid which was under increased pressure, but negative on examination. Ventricular air injection was interpreted as negative. Electroencephalogram suggested a spike and wave pattern, the waves being irregular and the rhythm abnormally slow.

The convulsions were partially controlled with phenobarbital and paraldehyde, and consciousness was regained over a 3 day period. The tremor and jerking movements of the right arm persisted but the right sided weakness improved. Generalized convulsions recurred on January 28, followed by an exacerbation of the myoclonic movements in both arms and 3 days of unconsciousness. Following this the pupils failed to react to light, the child was very dull and appeared to be blind. Death on March 19, 1946, followed a febrile episode. This was 87 days after admission. Numerous medications were without significant benefit.

Autopsy: These findings are taken from the autopsy protocol and were not witnessed by the present writer. The autopsy was performed on the day of death.

The body weighs 15½ kgs. and is 120 cms. long. The nutrition is poor. No macroscopic lesions are seen in the viscera.

The brain is fixed in the same manner as that used for the first case. The pia is delicate and can easily be removed from the brain substance. The vessels at the base and the dural sinuses are normal. The cranial nerves appear normal. The convolutions are flattened, but there is no pressure cone. On section the cut surfaces show normal structure grossly and the grey and white substance are sharply demarcated. The lentiform nucleus contains a few more whitish fibres than usual.

Microscopic Findings: The lungs contain minute foci of lobular pneumonia.

The liver shows fresh necrosis and fatty infiltration about the central veins. The kidney pelves and bladder show slight chronic inflammation. The optic nerve, retina and choroid of both eyes are normal. The remaining microscopic abnormalities are confined to the brain.

The material available for review of the central nervous system is limited to sections made from 11 paraffin blocks. Seven sections contain cerebral cortex, 3 of which show hippocampal gyrus and one of which shows the anterior end of the calcarine fissure. One section is through the putamen and pallidus. Another section shows the floor of the third ventricle just anterior to the mammillary bodies, some paraventricular nuclei and the anterior tip of the thalamus. Two sections include cerebellum and dentate nucleus. Half the medulla, including the inferior olive is on one block and there is a block of cervical cord.

Most of the sections of cerebral cortex are normal except for changes in some of the ganglion cells. These consist of chromatolysis, slight swelling, vacuole formation and pyknosis of nuclei. One section of hippocampus shows an area of moderate ganglion cell loss in Sommer's sector of the cornua ammonis. Many of the remaining cells about this area show poor staining qualities, chromatolysis, shrinkage or loss of nuclear outline. Some cells are represented by formless streaks or shadows. Also in the region of the cornua ammonis in two sections laminar areas of spongy rarifaction are seen adjacent to the ganglion cell layers. These areas show no evidence of gliosis or cellular reaction, and may represent artefact.

The cerebellar cortex shows rather marked loss of Purkinje cells. Many of those remaining show shrinkage, pyknosis, karyolysis, or even partial cytolysis. In some areas the molecular layer appears narrowed. Both sections of dentate nucleus show an appearance somewhat similar to that in the first case of this report. There are numerous vacuoles giving a somewhat spongy appearance to the tissue (Fig. 9). This change is concentrated at the junction between gray and white matter on both internal and external aspects of the nucleus. The ganglion cells are present in normal numbers, but some of them are severely altered. Swelling, cytoplasmic vacuoles, displacement of the nucleus, karyolysis and cytolysis is observed. As stated above, many of the ganglion cell changes may represent post mortem alterations. There is some increase in mature astrocytes in the dentate nucleus. There is no evidence of demyelinization in Weil stains of the cerebral cortex, cerebellum or spinal cord. There are no other lesions seen. Intracellular or extracellular inclusion bodies are not found. An iron stain including the pallidus is negative. The periodic acid routine fails to show glycogen deposits.

DISCUSSION

One is struck by the many similarities in the histories of these two siblings, and perplexed by the dissimilarity of the lesions. However, one must hasten to point out that the material now available for histological study in the case of G. T. is limited, there being no adequate sections of occipital cortex, thalamus, mid brain and pons. However

as opposed to the extensive lesions in M. T., only lesions in the dentate nucleus, cerebellar cortex and minimal changes in the gray matter of the cerebral hemispheres are demonstrated in G. T.

In searching through the histories to elucidate these differences, much remains obscure. The presence of ataxia and an intention tremor in the case of M. T. 10 months before death would suggest that the cerebellar lesions were present at that time. Perhaps the hyperreflexia and doubtful Babinski toe sign on the right side would indicate pyramidal tract involvement then as well. G. T. was not examined at a corresponding interval before death. M. T. survived for 59 days longer than G. T. after hospitalization became necessary, but it is questionable whether this interval contributed greatly to the more extensive demonstrable lesions in the former. That G. T. had more widespread lesion than were discovered is suggested by the report that he was thought to be blind and his pupils did not react to light during the final part of his illness. There was apparently little difference between the two cases in the severity of the generalized convulsions or of the myoclonic jerks.

Rather diverse pathological findings have been reported in myoclonus epilepsy, and the correlation betwen symptoms and lesions is still not entirely clear. The lesions in M. T. match in severity and extensiveness the most marked lesions described in the literature (3). It is safe to say that G. T. is much less severely affected. One does note that the dentate nucleus is involved in both cases, a point emphasized by Hodskins and Yakovlev (4). Intracellular inclusions were not found in either of the above cases, these bodies being frequently noted, but not invariably observed or specific for myoclonus epilepsy (5, 6). Deposits giving the staining reactions of glycogen have been noted in several cases of myoclonus epilepsy (7, 8). The method used failed to demonstrate glycogen in these cases. Iron deposits, especially in the pallidus, substantia nigra, and dentate nucleus are reported (9), but were not seen in these two instances.

From the pathological viewpoint the two cases under consideration might be considered as primary gray matter degenerations if one disregards a lesion in the case of M. T. in the medullary substance of the cerebellum. The destructive process, however, is more severe in M. T. than it is in the cases of degeneration of the cerebral gray matter

reported by Somoza, Alpers (10) and Freedom (11). Examination of the sections of the case described by Ford (12) shows a process in the cortical gray matter very similar to that described for M. T. The cerebellum was also involved, but the basal ganglia showed no definite alterations in this case. Myoclonus was not noted in any of these cases although other hyperkinetic manifestations such as choreiform and athetoid movements were recorded, as well as epileptiform seizures and mental retardation.

The similarities between the lesions in M. T. and the central nervous system lesions reported several days after exposure to severe anoxia (13, 14) are striking enough to bear examination. Animal experiments with anoxic anoxia (15) or the anemic anoxia of carbon monoxide poisoning (16) also demonstrate this similarity. The destruction of gangion cells or even complete softening, the demyelinization, the prominent microglial response and astrocytic proliferation are features of her lesions which are reported when longer intervals elapse following anoxia. The distribution of these changes is also compatible. The patchy involvement of the cerebral cortex with a tendency to be laminar, the involvement of the basal ganglia including the thalamus in some instances, as well as the cerebellum and some foci of gray matter in the brain stem fits the distribution of lesions in M. T. very well. The fact that severe hypoglycemia causes lesions like those of anoxia might indicate that such lesions are not specific for one type of metabolic disturbance, although some authors feel that anoxia is the fundemental defect underlying these lesions also (17).

However, it is not apparent that this child suffered from severe anoxia unless one implicates her convulsive seizures. Seizures usually leave no recognizable lesions, although lesions are reported in convulsive disorders (18, 19) in the cerebral cortex, especially the cornua ammonis, the cerebellum, caudate, putamen, pallidus, and also in the thalamus (20). They are minimal by comparison with those under consideration.

In considering epileptogenous foci, Penfield and Erickson (21) cite cases where a more severe damage was demonstrated and considered to be the result of seizures originating at the involved area. They suggest not only that the metabolic demands of the implicated neurons may exceed the temporary circulatory increase during the seizures,

but that these neurons may sometimes die as a result of the process. Other interpretations for these findings remain as possibilities, and in the absence of adequate confirmation one hesitates to take a stand on this evidence.

Both children had histories of a difficult birth, and in G. T. artificial respiration was required for 30 minutes. The evidence that the lesions are recent and progressive is too strong to permit a consideration of birth anoxia or injury as an etiological agent.

Finding ourselves unable to incriminate anoxia as a cause of the lesions, we must for the present regard the lesions as effects of a cause that is not yet recognized, this cause acting in different degrees in different cases. This underlying factor, whatever it may be, possibly consists in some hereditary metabolic disturbance of nerve cells, as is thought to occur in amaurotic family idiocy.

The pathogenesis of the diffuse central necrosis of the liver seen in both cases is not clear. This lesion, which was obviously terminal, is not mentioned in the previously reported cases of myoclonus epilepsy. Fatty infiltration without necrosis is described in the case of Frigerio (22). There is no stage of Wilson's hepato-lenticular degeneration which is known to give this lesion (23). Central necrosis is not a usual finding in deaths associated with the common types of epilepsy (24), but may be seen occasionally without any obvious etiological factor on examining the sections from a group of such cases.

SUMMARY

The clinical and pathological findings in two cases of hereditary myoclonus epilepsy are reported. The similarity of the histories of these 2 siblings as opposed to the dissimilarity in the extent of the lesions is pointed out. The lesions of the more severely involved case resemble those reported following severe anoxia, but no conclusive evidence incriminating anoxia can be summoned.

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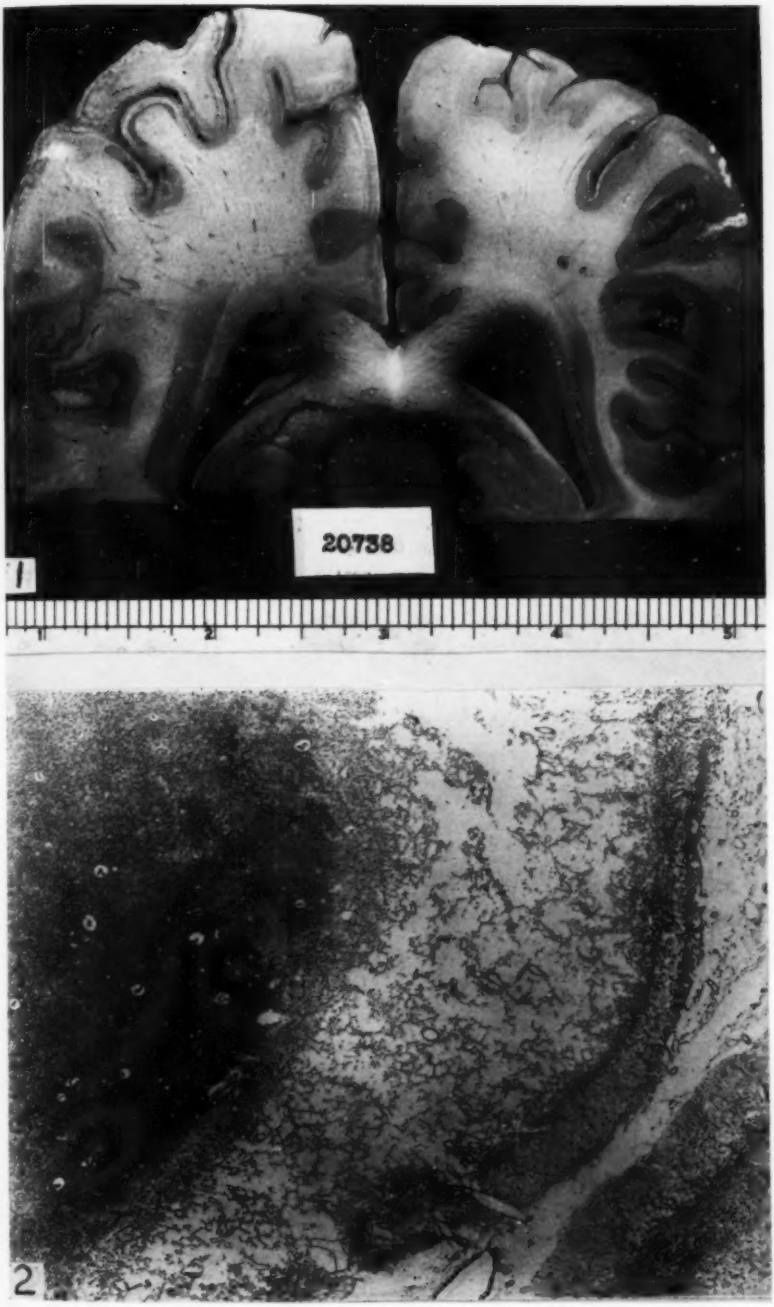
FIG. 1. Case M. T. Coronal section at the level of the splenium of the corpus callosum showing the thin cortical gray matter on the left side.

FIG. 2. Case M. T. Marked encephalomalacia of the cortical gray matter. Astrocytic hyperplasia is most marked in the molecular layer and at the junction of the gray and white matter. H. and E. ×20.



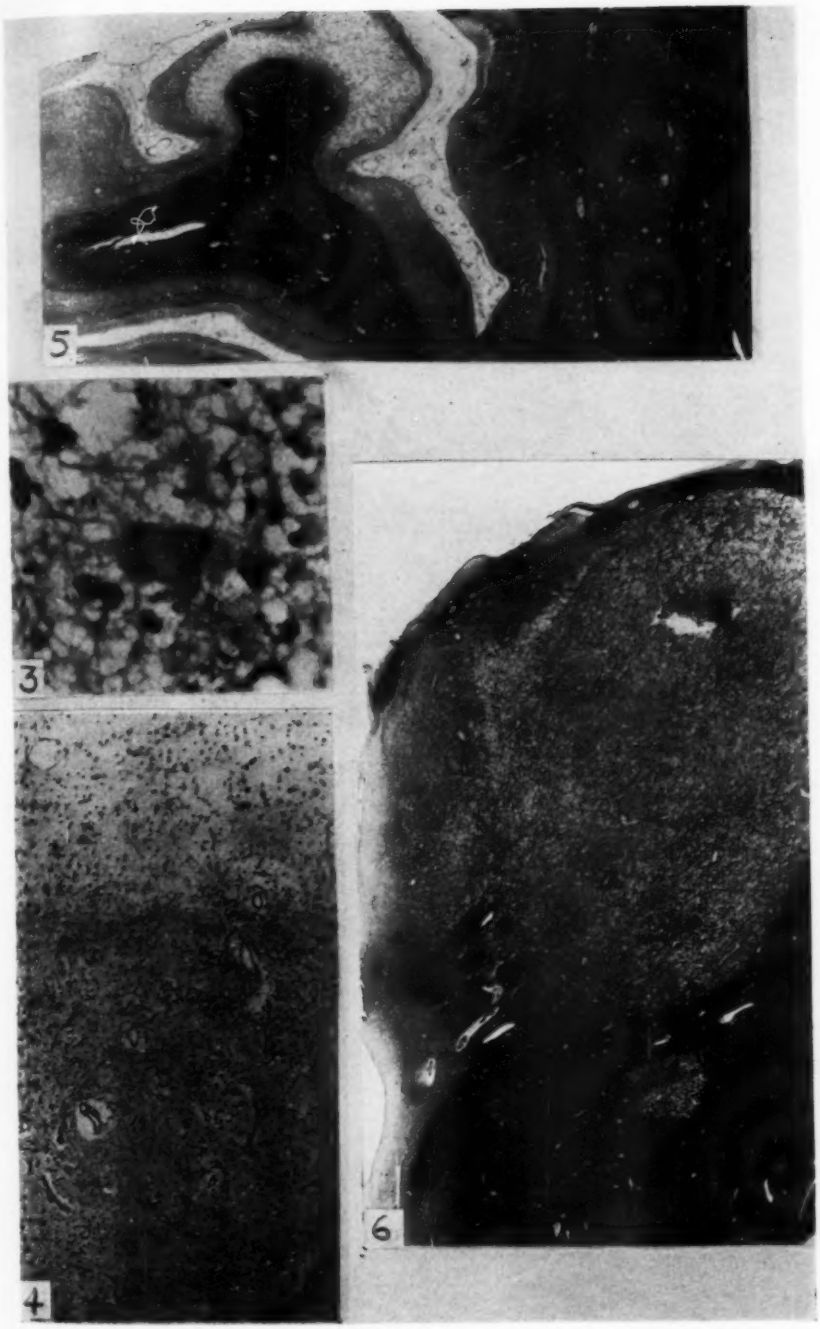
FIGS. 1 and 2

FIG. 3. Case M. T. Large binucleated astrocyte at the junction between the cortical gray and white matter. H. and E. ×500.

FIG. 4. Case M. T. Less extreme lesion showing the more superficial cortical layers. The few remaining ganglion cells are altered and the glial elements are more dense than normal. Toluidine Blue. ×60.

FIG. 5. Case M. T. Loss of myelin in the white matter underlying an involved area of cerebral cortex. Weil Myelin Sheath. ×3½.

FIG. 6. Case M. T. Status spongiosus involving most of the left thalamus. Weil Myelin Sheath. ×5.

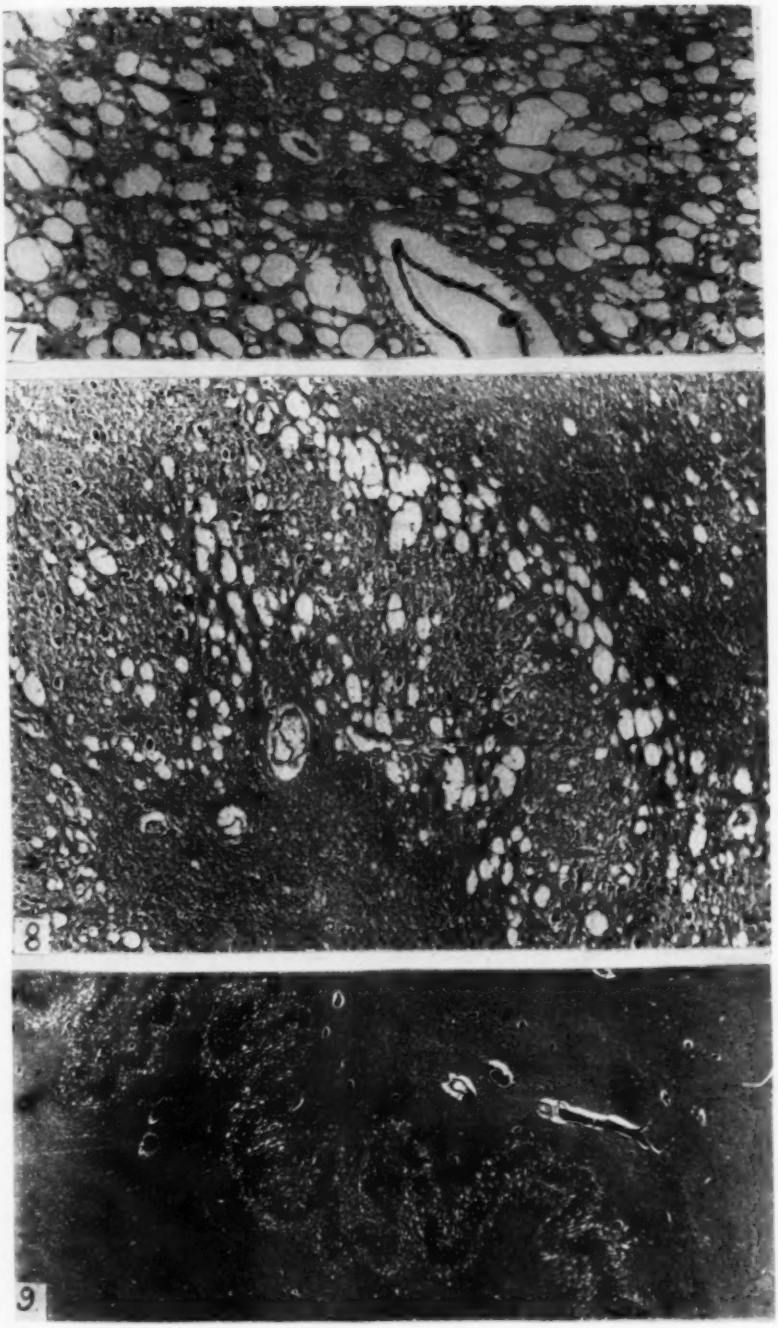


FIGS. 3, 4, 5, 6

FIG. 7. Case M. T. Status spongiosus of thalamus. A single surviving neuron may be seen in centre field. H. and E. ×100.

FIG. 8. Case M. T. Dentate nucleus, showing numerous large vacuoles chiefly localized at the junction between gray and white matter on both external and internal aspects of the nucleus. H. and E. ×60.

FIG. 9. Case G. T. Dentate nucleus, showing numerous vacuoles similar to those in the dentate lesion of M. T. H. and E. ×12.



FIGS. 7, 8, 9

OBSERVATIONS ON THE CLEARANCE METHOD OF DETERMINING RENAL PLASMA FLOW WITH DIODRAST, PARA-AMINOHIPPURIC ACID (PAH) AND PARA - ACETYL - AMINOHIPPURIC ACID (PACA)[1]

E. NEWMAN, A. KATTUS, A. GENECIN, J. GENEST, E. CALKINS AND J. MURPHY

WITH THE TECHNICAL ASSISTANCE OF MARION BIRMINGHAM AND MARGOT ROBINSON

From the Physiological Division, Department of Medicine, Johns Hopkins Hospital and University, Baltimore, Maryland

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In the course of an attempt to adapt to ambulatory subjects the clearance technique of determining renal plasma flow as originally devised by H. W. Smith and associates, it became evident to us that close scrutiny of the assumptions and methods involved was necessary. Our original purpose was to eliminate the constant infusion of diodrast, or para-aminohippuric acid (PAH), usually used to maintain a stable plasma concentration of the substance used for clearance determination, in order to be able to measure renal plasma flow on ambulatory subjects during different types of exercise. It would be convenient in such circumstances to be able to give a single injection of a substance and determine the renal plasma flow with a falling plasma concentration.

The observations to be presented were made because the clearances of diodrast and PAH in certain circumstances in man under basal conditions were neither constant nor independent of the plasma concentration. Some of the factors which might cause variation in the clearances were studied.

It is possible to determine glomerular filtration rate either with constant plasma concentration maintained by infusion or with falling plasma concentration after single injections of inulin, mannitol[2] or thiosulfate (1, 2). The fall in plasma concentration after single

[1] This work was supported by a grant from the Life Insurance Medical Research Fund.

[2] It is recognized that there has been recent evidence that there may be a small error in the mannitol clearance measurement of glomerular filtration rate.

injections of these substances follows a consistent logarithmic relationship with time, so that the plasma concentration for calculation of clearance can be obtained graphically from the line plotted from a few plasma analyses. With the subject in a basal state, the clearances of inulin, mannitol or thiosulfate are constant and independent of the plasma concentration, whether or not the concentration is held at any constant value or varies continuously throughout the clearance determination.

It is necessary for the validity of the clearance determination of renal plasma flow that the clearance be independent of the plasma concentration. The representation given by H. W. Smith et al. (3) of the relationship between plasma clearance and plasma concentration of a substance used for determination of renal plasma flow is described in Figure 1. Ideally, there exists a range of plasma concentration where the clearance is maximal and constant and is identical with the renal plasma flow. This relationship as represented implies that the clearance is constant and independent of concentration whether the plasma concentration is constant or varying within the maximal range.

"*Independence*" of plasma concentration can be defined in several ways. First, the clearance may be considered to be independent of plasma concentrations maintained at constant but different levels. This may be demonstrated by stepwise variation of the plasma concentration either upwards or downwards, determining the clearance while the concentration is held temporarily constant (*constant concentration—stepwise variation*). Secondly, the clearance may be termed independent of a *continuously changing plasma concentration*. "Independence" may be qualified in terms of the direction of the change in concentration: (*continuously rising or continuously falling concentrations*). Thirdly, the magnitude of the *range of plasma concentration* throughout which the clearance is independent may be different for each substance. This range will be limited by technical as well as physiological factors. At higher plasma concentrations there will be physiological self-depression of the clearance; at very low plasma levels the methods of analysis may be inaccurate.

If the plasma concentration is elevated sufficiently to cause depression of clearance, and then lowered so that clearance rises to its original or maximal value, another type of "independence" is demonstrated.

The *depression* of clearance is then said to be *completely reversible*, and hence the clearance at any given plasma concentration is *independent of previous plasma concentrations*.

Failure of the clearance to be "independent" when the plasma concentration is changed by any of the above methods may be due either to a real change in the renal plasma flow, or merely a change in clearance due to factors other than the renal plasma flow. In the first instance, the clearance is still a valid measure of renal plasma flow; in the second instance, the measurement is not valid.

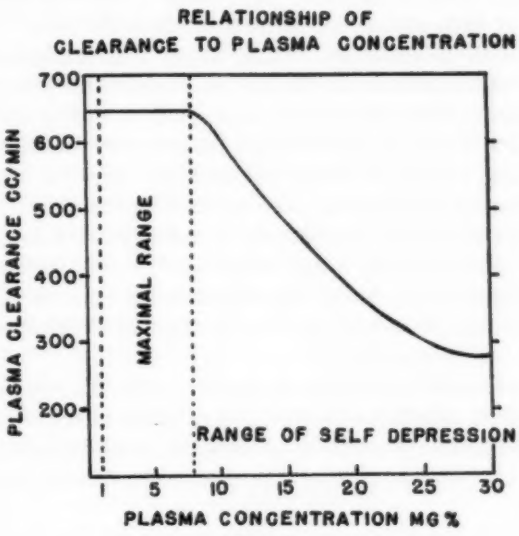

FIG. 1

It is conceivable that the substance injected itself may change the renal plasma flow, and that the magnitude and direction of the change depends on the plasma concentration. Either the injected substance or the simultaneously administered solutes could cause variations in body hydration, which may in turn alter the renal plasma flow. In ideal circumstances, the *actual* renal plasma flow should be independent of the plasma concentration of the substance injected, and salt, glucose and water injected with the substance ought to have no effect on renal hemodynamics.

There are several hypothetical circumstances which could produce invalid clearance measurements of renal plasma flow. One of the important assumptions involved in the measurement is that there is nearly complete[3] extraction of the substance from the plasma of low concentration flowing through the kidney. If the extraction is not complete, but is a constant fraction of the amount reaching the kidney, the clearance would not be valid quantitatively, but would change in direct proportion to the renal plasma flow. Incomplete extraction or failure of the renal tubules to remove all of the substance flowing through the kidney is demonstrated clearly by self-depression of the clearance at high plasma concentrations when the renal tubules are *overloaded* with the substance. Other causes of incomplete extraction, when the tubules are not overloaded, may be either a *primary failure of the tubular secretory mechanism* or the interference by another substance which inhibits or competes for the secretory mechanism (*secretory blockade*). Such a competing substance may be some of the original compound which has been modified in the body.

Indirect evidence for establishing complete extraction can be obtained by demonstrating a high clearance (i.e. that clearance value which is highest most nearly approximates the renal flow, assuming that falsely high clearances due to manufacture of the substance by the kidney do not occur).

Direct evidence for establishing complete extraction can be obtained by analysis of simultaneous arterial and renal vein plasma. However, demonstration of complete extraction by renal vein plasma analysis is not complete proof that the clearance measures renal plasma flow.

Chemical alteration of the substance might take place in the kidney so that its presence in the urine or in renal vein plasma would not be detected by the usual method of analysis. Complete proof that a substance measures renal plasma flow would require the demonstration of complete extraction, and, in addition, that the extracted substance was collected *completely* and *simultaneously* in the urine in a measurable form.

Here two secondary factors may possibly cause error and invalid

[3] Complete except for a small amount of blood which goes through non-secretory tissue of the kidney.

measurements. The first is the possible discrepancy between arterial and venous plasma concentrations. With a falling plasma concentration, arterial concentrations would be lower than those obtained from the brachial veins since the tissues of the arm would be contributing their store to the circulation. The magnitude of the gradient between arterial and venous concentrations might vary with the rapidity of the fall in arterial concentration and with the rate of diffusion of the substance from tissues to capillary blood. Since the kidney would receive the substance from arterial plasma, any discrepancy between arterial and brachial venous plasma concentrations would cause the clearance calculation to be falsely low.

The second factor interfering with simultaneity of plasma concentration and the collection of the urine is the *delay time*. In all probability the delay time is entirely the time required for the urine leaving the collecting tubules in the kidney to traverse the calyces, renal pelvis and ureters to the bladder. With a falling plasma concentration a significant delay time would result in a falsely high clearance calculation because urine which was formed at a higher plasma concentration would be collected simultaneously with plasma at a later lower concentration. It is conceivable also that the delay time might change significantly during the experiment due to increase or decrease in urine flow. A decreasing urine flow would result in a storage of some substance in the urinary tract and a fall in clearance calculation; conversely, an increasing urine flow would release some of the stored substance and increase the clearance.

METHODS

(1) *Serum and Urine Determinations*

Mannitol, inulin and PAH[4] were determined in serum or heparinized plasma and urine by the methods suggested by Goldring and Chasis (4), with the exception that precipitation of proteins was carried out by Somogyi zinc sulfate-sodium hydroxide instead of by cadmium sulfate reagents. Diodrast was determined by the method of Alpert (5) using this same method of precipitation of plasma proteins.

[4] The following abbreviations will be used: Diodrast—D; Para-amino-hippuric acid—PAH; Para-acetylaminohippuric acid—PACA; the conjugated product of PAH formed in the body—"PACA".

The determination of PACA or of PAH conjugated in the body ("PACA") required some investigation before complete recovery of PACA was obtained. PACA was determined as PAH after hydrolysis, and the concentration of PACA was calculated from the difference in PAH concentration before and after hydrolysis in the same specimen. Heat and acid required for hydrolysis may cause loss of PAH. The method finally developed is a slight modification of Smith's (9), as follows:

Ten cc. of protein-free filtrate or diluted urine containing not more than 0.04 mg. nor less than 0.01 mg. of PAH are placed in a test tube[5] and 2 cc. of 1.2 N hydrochloric acid are added. About 0.5 cc. of melted paraffin is dropped on top of the fluid in each of the tubes. The tubes are placed in a boiling water bath for 90 minutes with a rubber stopper loosely inserted. The tubes are then cooled and rotated so that the paraffin sticks to the walls. Addition of the usual amounts of nitrite, sulfamate, and coupler is then carried out in the test tubes for the determination of PAH. It is unwise to add acid to the tubes for the determination of free PAH unless the whole determination is completed immediately, because conjugated PAH in urine may be partially hydrolyzed by standing at room temperature with acid added.

While it is likely that the conjugated PAH found in human serum and urine after the injection of PAH is acetylated PAH, as far as we know it has not been definitely identified and shown to be the only conjugate produced. While the method of determination has been shown to be accurate for PACA, it is assumed to be valid also for PAH conjugated in the body.

Blank determinations for diodrast, PAH and PACA were made on serum and urine obtained before the administration of these substances. No blank substances for diodrast were found in human serum. The blank serum determinations for PAH and PACA varied from 0 to .05 mg.% and were subtracted from subsequent determinations. Urine blank determinations for all substances were made in order to rule out the possible presence of interfering substances such as sulfonamides. The values for urine blank substances are negligibly small in the final calculations of clearance. The serum blanks for

[5] Thin walled, 16 mm. inside diameter, and 220 mm. long.

mannitol, thiosulfate and inulin were always determined and subtracted from subsequent values.

Creatinine was determined on tungstate filtrates of serum and on diluted urine. Serum proteins were precipitated by adding 2 cc. of serum to 22 cc. of water, then 8 cc. of 5% sodium tungstate and 8 cc. of 1/3 N H_2SO_4 solution. To 10 cc. of the serum filtrate or of diluted urine, 5 cc. of alkaline picrate solution was added and the color read with a photoelectric colorimeter (520 μ filter.) (Alkaline picrate solution, made fresh and used within five minutes after preparation, contains one part of 10% NaOH and five parts of picric acid solution containing 11.75 gm. of picric acid per liter.)

(2) *Preparation of Para-acetylaminohippuric Acid for Parenteral Use*

A 20% solution of sodium salt of PAH from ampoules[6] is diluted with an equal volume of pyrogen-free distilled water. Acetic anhydride in slight excess of the ratio of two mols to one of PAH is added while stirring, and the solution is allowed to stand at room temperature for 30 minutes with occasional shaking. The mixture is cooled in an ice bath and filtered by suction through a Buchner funnel. The precipitate is washed several times with ice-cold pyrogen-free distilled water, then with 95% ethyl alcohol, and spread out on dry filter paper to dry in room air overnight. Further drying can be carried out in a desiccator. The precipitate should be protected from light during the drying and stored in a dark bottle.

The substance causing the yellow color sometimes seen in PAH solutions is completely removed by the acetylation since it appears in the washings and the resultant precipitate is white. The yellow color in PAH solutions is produced by some unidentified impurity—possibly an oxidation product.

For intravenous administration, a 5 or 10% solution of PACA was made by dissolving the PACA in NaOH until the pH of the solution was approximately 6.0. (This requires roughly 5 cc. of 0.75 N. NaOH per gram of PACA.) The solution can be sterilized by filtration or by autoclaving. Solutions of PACA made in this way are free of PAH if filtered, but show traces (0.1%) of PAH if autoclaved.

The solubility of PACA at varying pH's is important because it is

[6] We are greatly indebted to Sharp and Dohme, Philadelphia, for generously supplying ampoules of PAH and mannitol for parenteral use.

conceivable that in acid urine crystalluria might occur. The solubility of PACA was determined. At pH 5.0 in water at room temperature the solubility of PACA was 6.0 grams/100 cc. Within the physiological range of urine pH, precipitation should not occur in urine containing concentrations which are obtained during clearance studies with low plasma concentrations and an adequate urine volume.

(3) *Clinical Procedure*

Patients were examined in the fasting state in bed. Single injections of substances were given by syringe at the rate of 4 cc. per minute. About 20 minutes after the end of the injections, plasma and urine collections were started. Constant infusions were maintained by a gravity drip regulated by a tunnel clamp on the tubing. The rate of drip was regularly and frequently counted and recorded to assure constancy. Urine flow was kept as high and as constant as possible by administration of water by mouth to minimize delay time or storage in the urinary tract.

For calculation of clearances with a falling plasma concentration, the concentration at the mid-time of the urine collection period was used. This was obtained from a graph of the plasma concentrations which yields nearly a straight line on semi-log paper (e.g. Figure 6 and Figure 11). No correction for delay-time was made in any of our calculations. Application of a delay-time correction to any of the falling clearances does not alter significantly the direction or magnitude of the change in clearances. Also no attempt was made to introduce corrections for changes in urine flow since diuresis was maintained.

Metycaine was used as a local anesthetic whenever necessary Unlike procaine, it does not interfere with PAH determination. Preventing the anticipation of possible pain on repeated vein punctures may be important in maintaining equanimity during the clearance procedures. Urine was collected by catheter in hospital patients and the bladder washed with water and air to insure accurate collections. Normal subjects were allowed to stand to void. Other details of experiments are given with the charts and captions.

In dogs similar techniques were used. Creatinine was administered by mouth two hours before the experiments.

RESULTS

Observations on Diodrast

The first observations in man of a falling clearance occurring with falling plasma concentration were obtained after single injections of diodrast (Figure 2). These experiments raised the questions as to

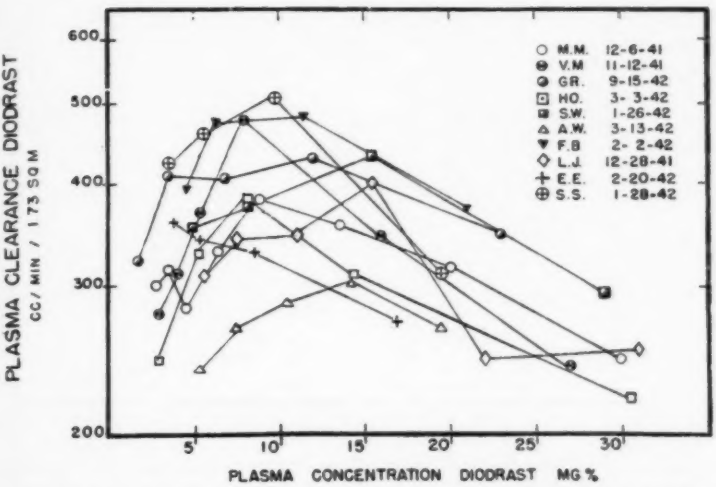

FIG. 2. The clearance of diodrast determined with falling plasma levels of diodrast after the single injection of a large amount in ten patients. As the plasma level fell from high levels, where there was depression of clearance, to the region of 10 mg. %, the clearance rose. But as the venous plasma level fell below 10 mg. % into the range where clearance should be maximum and constant, the clearance fell progressively.

whether the depression of the clearance at low plasma concentrations was caused by the continuous changing of the concentration or whether the depression of clearance was caused by the exposure of the tubules to a previously higher concentration. Accordingly, we attempted to determine what effect exposure of the kidney to higher plasma concentrations would have on the clearance determined with constant plasma concentrations.

In Figure 3 is the result of temporarily raising the diodrast plasma

concentration and then returning to a low sustained level. The clearance was slightly self-depressed when the plasma concentration was raised from 1.0 to 8.0 mg.%, but was further depressed when the

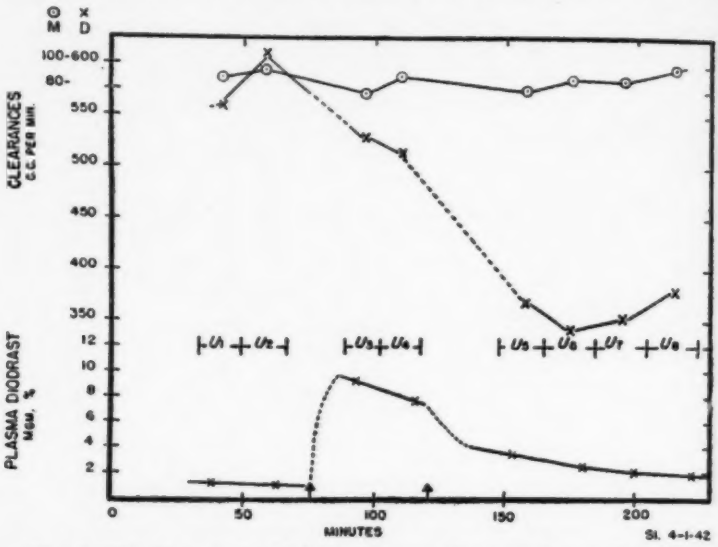

FIG. 3. The effect on the diodrast clearance of a normal man of raising and lowering the plasma diodrast concentration by continuous intravenous infusions. Simultaneous mannitol clearances were also determined.

Patient, a 17 year old male, height 71 inches, weight 142 lbs.

⊙ = Mannitol clearance

× = Diodrast clearance (above) and plasma concentration (below)

Protocol: Time = 0 minutes—Prime 50 cc. of 50% mannitol and 1 cc. of 35% diodrast solution; Time = 10 minutes—Sustaining infusion begun containing 3 cc. of diodrast and 37 cc. of mannitol solution in 150 cc. saline at 2 cc. per minute; Time = 70 minutes—Prime of 9 cc. of diodrast solution given and sustaining infusion changed to infusion containing 27 cc. of diodrast and 37 cc. of mannitol in 150 cc. of saline at 2 cc. per minute; Time = 120 minutes—Sustaining infusion changed to solution used originally during time 10 minutes to 70 minutes.

plasma concentration returned to a sustained low level. The mannitol clearance remained constant throughout the procedure, indicating that the patient was in a steady state and that the urine collections were accurate.

A similar observation on another patient is recorded in Table I. Elevation of the plasma diodrast concentration to 8.0 mg.% caused no self-depression of clearance in this patient. In this case the infusion was stopped abruptly. The clearance determined later while the plasma concentration was falling slowly was markedly depressed.

After these observations, the study was interrupted for a few years and the use of diodrast was abandoned for para-aminohippuric acid, which is determined more easily chemically.

TABLE I

The Effect of Raising and Lowering the Plasma Concentration on the Clearance of Diodrast in a Patient

TIME	PLASMA CONCENTRATION	CLEARANCE DIODRAST	URINE FLOW	DURATION OF URINE COLLECTION
Mins.	*mg.%*	*cc./min.*	*cc./min.*	*Mins.*
22	2.18	515	5.4	19.3
44	1.42	522	10.8	18.3
80	8.3	503	11.2	16.5
98	7.5	554	11.1	21
132	3.4	425	10.5	16.2
150	2.8	290	11.0	16.0

Protocol: At time = 0 minutes—Prime 1 cc. of 35% diodrast and infusion begun containing 3 cc. of diodrast in 150 cc. saline at 2 cc. per minute; At time = 55 minutes—Prime 9 cc. of diodrast and infusion changed to solution containing 27 cc. of diodrast in 150 cc. saline at 2 cc. per minute; At time = 110 minutes—Infusion stopped. Patient M. M., 12/13/41, height 67 inches, weight 135 lbs.

Observations on PAH

Single injections of PAH in several individuals also produced falling clearances (Figure 4). The plasma concentrations initially were not as high as those obtained with diodrast injections (Figure 2), so that the self-depression of clearance at high concentration is not illustrated. A fall in clearances occurred throughout the range of plasma concentration where the clearance is thought to be maximal and constant.

Lack of reversibility of PAH clearance to original high normal value after temporary elevation of the plasma concentration can also be demonstrated (Table II). The clearance remains low even though

the plasma level is sustained by a constant infusion at a lower level than used previously when the clearance was maximal.

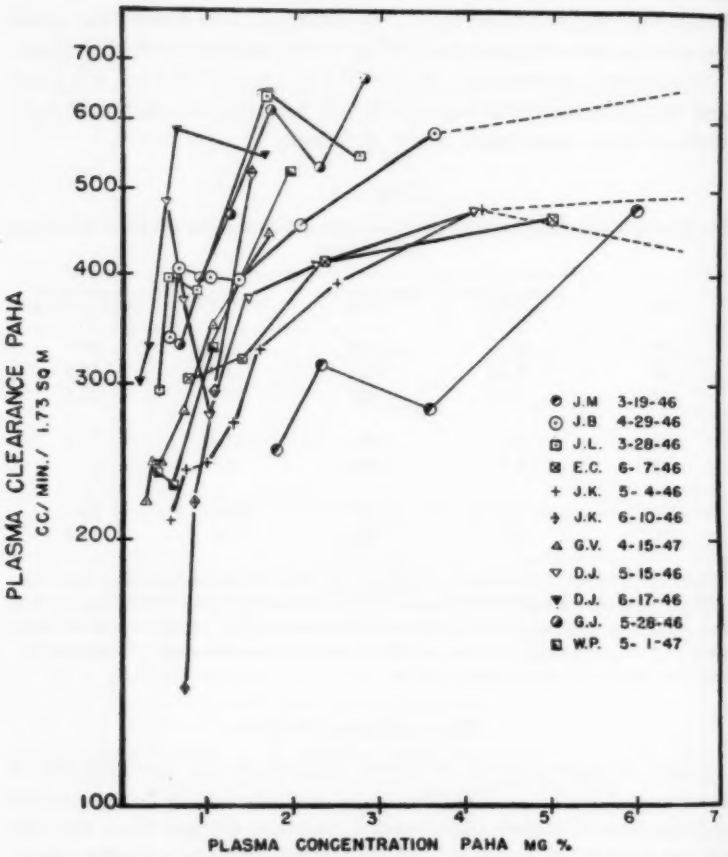

FIG. 4. The clearance of PAH falls as the plasma concentration falls after single injections in eleven subjects

The behaviour of the clearance of PAH was observed in dogs given single injections. In the dog, the clearance of PAH does *not* fall when the plasma concentration falls throughout the low range (below 8

mg.%), where clearance theoretically should be maximal (Figures 5 and 6). Thus, the PAH clearance behaves differently in man and dog after single injections.

TABLE II

Depression and Lack of Reversibility of PAH Clearance during Constant Infusion Due to Temporary Elevation of Plasma Level

Normal Male Subject, J. M.; Age 25; Surface Area 1.9 Sq M.
Date: 6/5/46.
Calculated normal plasma clearance 760 cc./min.

TIME	PROCEDURE	PAH PLASMA CONC. MID-PERIOD	PLASMA CLEARANCE PAH	URINE FLOW
		mg./100 cc.	*cc./min.*	*cc./min.*
8:03	I.V. drip PAH 8 mg./min. started			
8:06	2 cc. PAH 20% I.V. prime			
8:16	Urine discard			
8:30	U_1	1.95	590	4.55
8:45	U_2	1.52	730	13.8
8:51–8:56	15 cc. PAH 20% I.V. Drip PAH discontinued			
9:07	Urine discard			
9:24	U_3	12.44	381	15.5
9:44	U_4	5.1	252	18.5
10:04	U_5	2.4	460	15.3
10:24	U_6	1.46	436	6.0
10:27	I.V. drip PAH 8 mg./min. started			
10:40	U_7	0.97	410	7.9
11:04	U_8	0.83	464	9.5
11:24	U_9	0.90	454	12.5
11:44	U_{10}	0.96	470	12.3
12:04	U_{11}	0.97	496	9.2

The effect of temporary elevation of the plasma concentration of PAH upon the clearance determined at low sustained concentration was not investigated in the dog.

The constancy of PAH clearance when the plasma concentration is

kept level by a sustaining infusion over long periods has been demonstrated many times before. In Table III are PAH clearances in one of our subjects over the course of three and three-fourths hours.

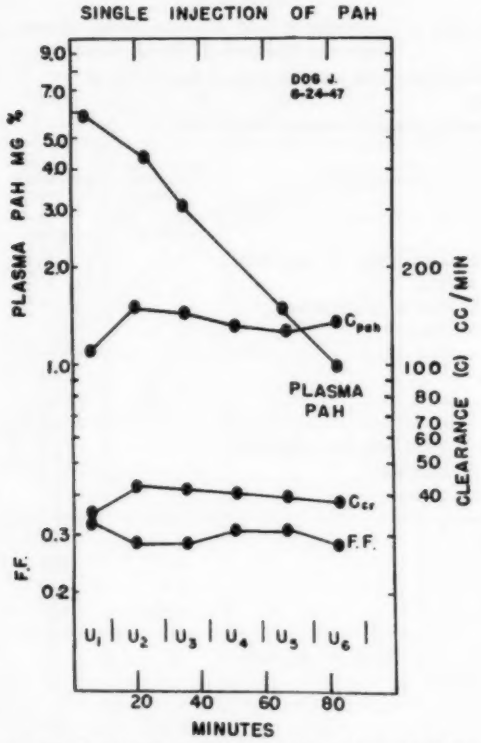

FIG. 5. Constancy of PAH clearance (C_{PAH}) in a dog with falling plasma concentration after single injection of 5 cc. of 20% solution of PAH in 20 cc. of saline. C_{Cr} is clearance of creatinine.

Thus, exposure of the kidney to the same plasma concentration for many hours did not cause significant change in clearance.

The concept that the falling clearance of PAH in man with falling plasma concentration was caused by the venous plasma not being representative of the arterial blood was explored. If the depression of

clearance were due to a high venous concentration, arterial plasma determinations should reveal an arteriovenous difference, and the clearance calculated from arterial concentrations should be constant.

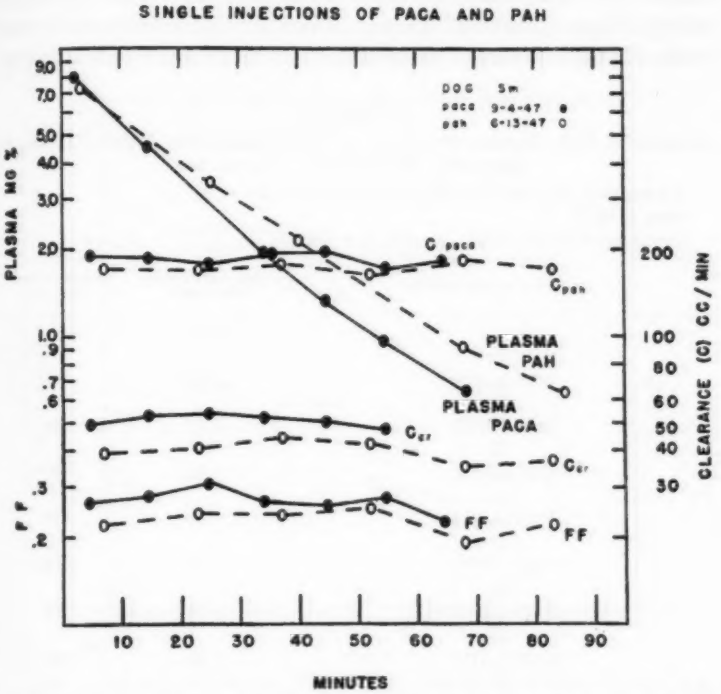

FIG. 6. Single injections on different days in the same dog of 1.0 gram of PAH in 20 cc. saline on 6–13–47, and 1.0 gram of PACA in 30 cc. water at pH 7 on 9–4–47 show constancy of clearance with falling plasma concentrations over the same range for both substances.

Symbols: C_{PAH} is clearance of PAH
C_{PACA} is clearance of acetyl PAH
C_{Cr} is clearance of creatinine
F.F. is filtration fraction

In Figure 7 an arteriovenous difference in PAH plasma concentrations is demonstrated, but the falling concentration curves are parallel so that the "arterial" clearance falls. Thus, after a single injection of

PAH in man, the venous concentration is higher and proportional to the arterial plasma concentration. The dog shows a constant clearance of PAH determined from venous plasma (Figures 5 and 6), so that it is unlikely that an arteriovenous difference would explain the falling clearance obtained in man. Evidence that PAH is rapidly and freely diffusible from plasma to tissues will be presented later, indicating that it is unlikely that PAH would be stored in the tissues and released to the venous blood at a later time.

TABLE III

Constancy of PAH Clearance Over Three and Three-fourths Hour Period of Continuous Intravenous Administration of PAH in Man

Normal Male Subject, J. B.; Age 29; Surface Area 2.1 Sq M.
Date: 6/14/46.
Continuous I.V. drip PAH. Started at 8:00 A.M.

URINE COLLECTION PERIODS	PAH PLASMA CONC. MID-PERIOD	PLASMA CLEARANCE PAH
	mg./100 cc.	*cc./min.*
U_1 8:15 to 9:17	1.02	608
U_2 9:17 to 9:34	1.45	516
U_3 9:34 to 9:55	1.42	407*
U_4 9:55 to 10:14	1.40	518
U_5 10:14 to 10:33	1.39	506
U_6 10:33 to 10:53	1.35	579
U_7 10:53 to 11:13	1.23	582
U_8 11:13 to 11:34	1.19	535
U_9 11:34 to 11:54	1.30	572

* Small amount of urine lost.

Observations on PAH Conjugated in Vivo ("PACA") and Para-acetylaminohippuric Acid (PACA)

The behaviour of the clearance of PACA in dog and man was investigated. The solubility of PACA within the range of pH in urine

was determined. Clearances of PACA were determined after single injections in dogs. In Figure 6 the clearance of PACA is shown to be identical with the PAH clearance and is constant throughout the falling plasma concentration from 8 to 1.0 mg.%. No self-depression of clearance occurred within this range. A larger dose of PACA was given in another experiment when the plasma concentration was raised

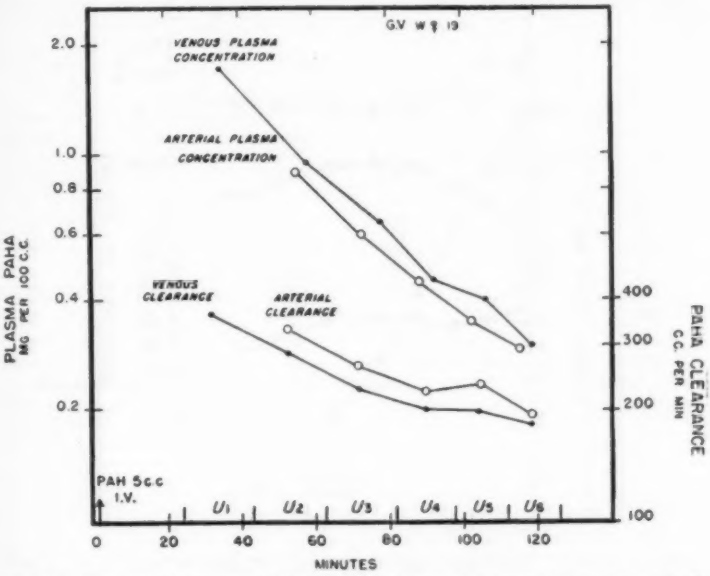

FIG. 7. The arterio-venous difference in plasma concentration after a single injection of PAH, and the clearance calculated from both plasma curves. The arterial blood was obtained from an inlying needle in the femoral artery and the venous blood from the arm vein. The clearances fall in a parallel manner.

to 28 mg.% (Figure 8). Self-depression of clearance was present until the plasma concentration fell to 8.0 mg.%, after which the clearance was maximal and constant. Thus, in the dog, PACA clearances like PAH clearances do not show depression or fall with a falling plasma concentration after either large or small single intravenous injections.

Similar data on a falling curve in a dog with an explanted kidney are given in Table IV with simultaneous determinations of renal vein

plasma concentration of PACA. The renal extraction of PACA was high throughout the whole range of plasma concentration. It appears, therefore, that PACA clearance in the dog measures the renal

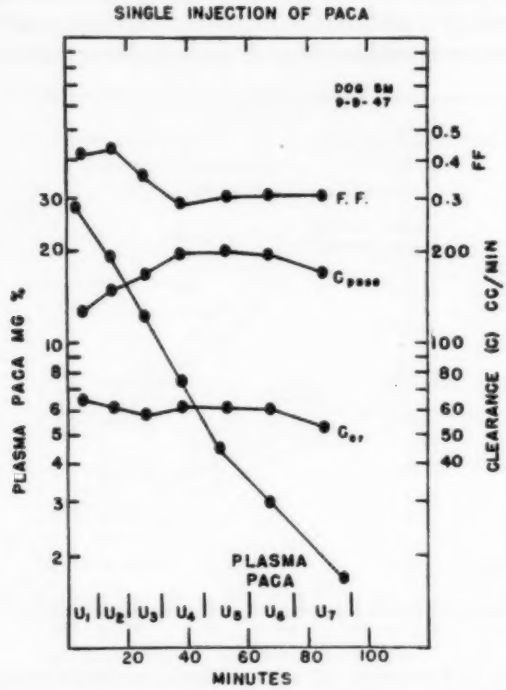

FIG. 8. Single injection in a dog of 4.0 grams of PACA in 55 cc. of water (pH 7). The ratio of creatinine clearance (C_{Cr}) to PACA clearance (C_{PACA}) is constant (F. F.) between plasma concentration of 8.0 to 1.6 mg. %. There is depression of PACA clearance above a plasma concentration of 8.0 mg. % so that the filtration fraction is falsely elevated (F. F.).

plasma flow as well as PAH clearance and that the clearance is independent of a changing or a previously high plasma concentration.

Determinations for the presence of "PACA" in plasma and urine after injections of PAH in the dog revealed no evidence of conjugation of PAH by the dog in any of the above experiments, nor was there any

evidence of de-acetylation of PACA, since no PAH could be found after injections of PACA.

Because of the difference in behaviour of PAH clearances in dog and man after single injections, and because the dog *does not* conjugate PAH whereas man *does*, it seemed important to determine the amount of conjugated PAH in man and its mode of excretion. The results in Figure 9 show that as the PAH clearance falls the clearance of "PACA"

TABLE IV

The Renal Extraction of PACA in a Dog

PERIODS	TIME	MID B PACA	C_{PACA}	$C_{CREAT.}$	F. F.
	Mins.	*mg.%*	*cc./min.*	*cc./min.*	
1	11.5	11.7	87.1	27	.31
2	23.0	7.3	108	31	.28
3	33.5	4 7	126	28	.224
4	50.5	3.25	112	30	.27
5	69.5	1.80	122	32	.26
6	85.5	1.11	107	32	.298
7	104.0	.76	88.5	30	.34

PERIPHERAL PACA	RENAL VEIN PACA	EXTRACTION
mg.%	*mg.%*	%
9.7	1.9	80
4.8	1.1	77
1.13	.12	90
.67	.10	85

Dog Gerry. Peripheral blood obtained from jugular vein. Renal vein blood obtained by direct puncture of renal vein in an explanted kidney preparation. Single injection of 1 gram of PACA in 20 cc. water at pH 7.

Explantation of kidney performed by Dr. Willard Goodwin, Resident Urologist.

remains higher. Fluctuation in the calculated clearance of "PACA" is seen because the plasma concentration of this substance is low and is obtained by difference between the PAH and total concentrations. Thus, the accuracy of the plasma "PACA" determination is questionable. Nevertheless, it seems significant that the clearances of conjugated PAH are always higher than those of PAH.

The proportions of PAH present in the urine during the observations in Figure 9 are given in Table V. As the rate of excretion and

the plasma concentration of PAH fall, there is an increased proportion of conjugated PAH in the urine.

In view of the fact that conjugation of PAH may have been responsible, at least in part, for the falling clearances, we have made some

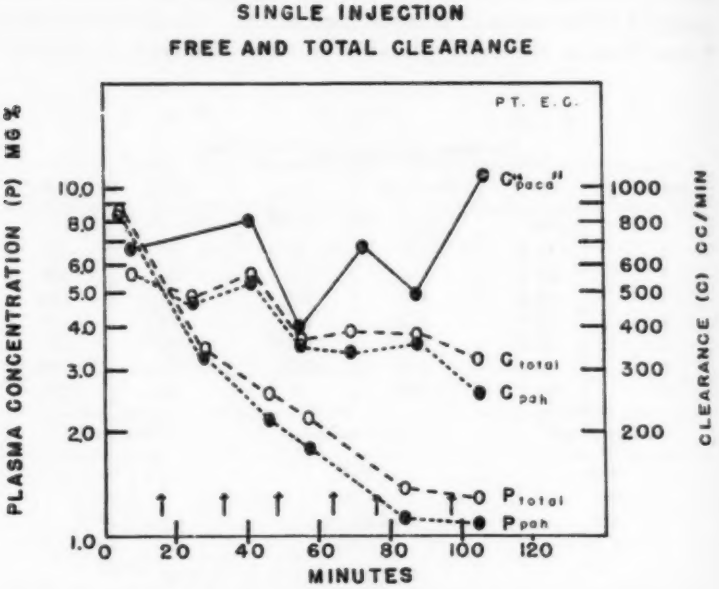

FIG. 9. The plasma concentration and clearances of PAH and "PACA" (PAH conjugated in vivo) after a single injection of PAH in a normal subject. The clearance of "PACA" is always higher than the clearance of PAH which steadily falls. Calculation of clearance from total concentrations (PAH + "PACA") also yields a falling curve. See Table V for the relative amounts of PAH and "PACA" in the urine in this experiment.

preliminary observations on the behaviour of PACA clearances after single injections. In Figure 10, 11 and 12 are observations, after single injections, showing that the clearance of PACA does not show the progressive fall encountered previously with PAH clearances within the range of plasma concentration of 7 to 1.0 mg.%. (Compare with PAH clearance in Figure 8.) Variations in PACA clearance

were accompanied by proportionate changes in the clearance of inulin so that the filtration fraction (F.F.) remained nearly constant. The constant F.F. confirms the fact that there is no progressive depression of PACA clearance. High filtration fractions were encountered in the cases with hypertension or cardiac failure.

TABLE V

The Relative Concentrations of "Free" PAH and Total PAH (Free + Conjugated) in Urine Following Single Intravenous Injection of PAH in Man

Normal Male Subject, E. C.; Age 21; Surface Area 1.83 Sq M.
Date: 12/23/47.
PAH—5 gms. I.V. between 9:33 A.M. and 9:46 A.M.

URINE COLLECTION PERIOD	PAH (UV)	TOTAL PAH + CONJUGATED PAH (UV)	CONCENTRATION RATIO	PAH PLASMA CONC. MID-PERIOD
	mg./min.	*mg./min.*	*PAH/Total*	*mg./100 cc.*
U_1 10:03 to 10:18	4.10	43.0	95.5%	7.3
U_2 10:18 to 10:36	1.73	18.8	92.0%	3.7
U_3 10:36 to 10:51	1.30	16.0	81.4%	2.43
U_4 10:51 to 11:06	6.57	8.21	80.0%	1.85
U_5 11:06 to 11:19	4.89	6.79	72.0%	1.46
U_6 11:19 to 11:40	3.97	5.10	77.9%	1.12
U_7 11:40 to 11:56	2.75	4.01	68.5%	1.08

See Figure 10 for the behaviour of the clearances of PAH and conjugated PAH.

After injections of PACA into man, no "free" PAH was found in the plasma or urine. Thus de-acetylation of PACA does not occur in man.

Concerning Priming Doses

It is possible that the temporary elevation of plasma concentration by the priming dose to levels far above the ultimate level to be maintained by the constant infusion, may cause a falling plasma level during the first periods of clearance determination, with resultant depres-

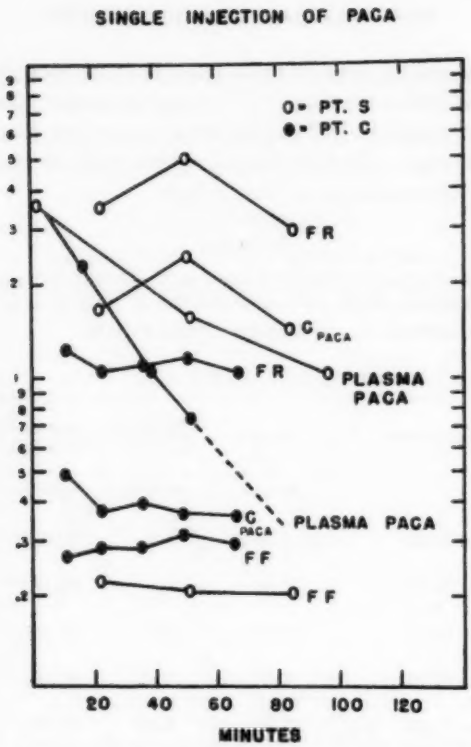

FIG. 10. The constancy of filtration fraction (F. F.) in two children with falling plasma PACA concentrations after single injections of thiosulfate or inulin and PACA.

Patient C (●). Diagnosis: Coarctation of aorta with hypertension.

Weight: 98.5 lbs., Height: 58 inches.

Single injections of 10 grams of sodium thiosulfate in 100 cc. water with 2.35 grams of PACA in 100 cc. water at pH 7.

Patient S (○). Diagnosis: Coarctation of aorta after surgical resection of aortic isthmus by Dr. Alfred Blalock. Blood pressure normal.

Weight: 37.7 lbs., Height: 42.3 inches, Age: 5 years.

Single injections of 3 grams of inulin in 30 cc. saline and 1.0 gram of PACA in 25 cc. water at pH 7.

While the filtration rate and clearance of PACA vary, particularly in patient S, there is constancy of the filtration fraction (F. F.). Marked variations in clearances with constant F. F. are probably due to inaccurate urine collections. There is a constant relation of the clearance of PACA to the clearance of thiosulfate or inulin with a falling plasma PACA concentration over the range of 3.6 to less than 1.0 mg. % in both patients. The higher F. F. (.27 – .30) is found in the child with hypertension. Urine was collected by catheter.

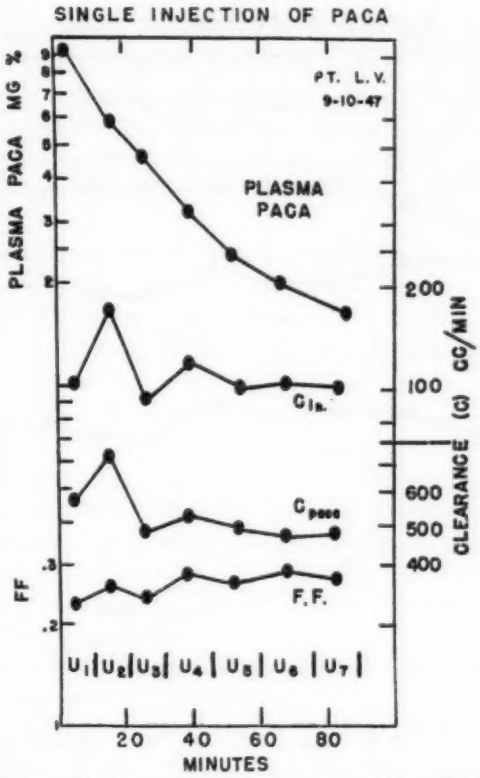

FIG. 11. Patient L. V., Age: 18, Weight: 141 lbs., Height: 68 inches, diagnosis: hypertension. Single injections of inulin 10 grams in 100 cc. saline and 5.0 grams PACA in 100 cc. water at pH 7.

The marked variations in clearance values in the first three periods are probably due to difficulty encountered in urine collection. The F. F. was within the range of .23 to .29 as the PACA plasma level fell from 9.28 to 1.65 mg. %. Apparently there was no significant self-depression of the PACA clearance at a plasma concentration of 5 to 9 mg. %, since the F. F. was not elevated. Depression of PACA clearance should raise the F. F.

sion of clearance, such as is demonstrated with falling plasma levels following single injection with no sustaining infusion, or may cause a depression of clearance due to temporary exposure to higher concentrations.

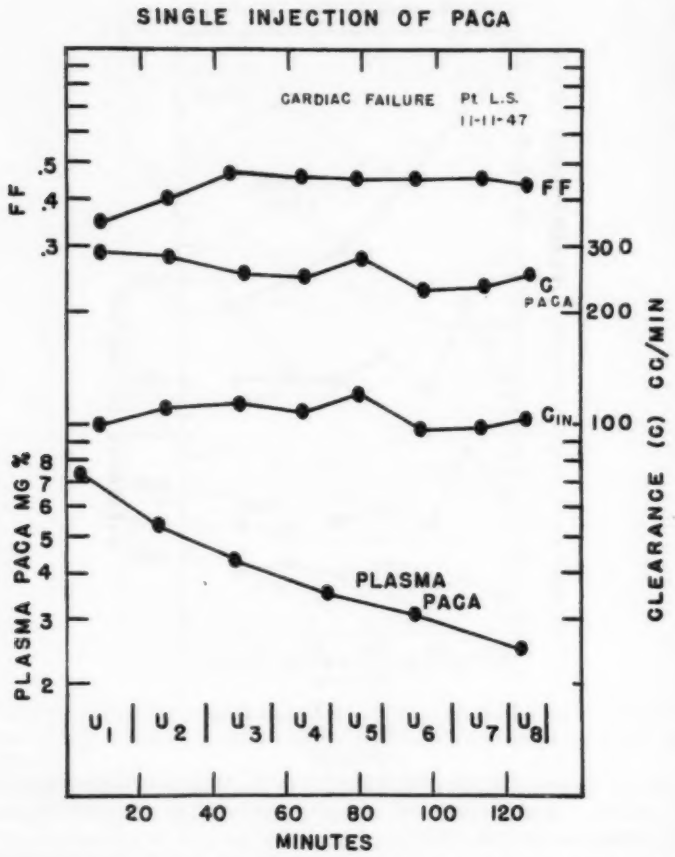

FIG. 12. Patient L. S., Age: 38. Cardiac failure due to rheumatic heart disease with murmurs of mitral stenosis and insufficiency and aortic insufficiency with peripheral edema, orthopnea and elevated venous pressure. Patient was propped up in a cardiac bed throughout the examinations.

Single injections of 10 grams of inulin in 10% solution and 5.0 grams of PACA in 50 cc. water at pH 7. Urine collected by catheter.

The filtration fraction is abnormally high, but after the first three periods it is constant with the plasma concentration of PACA falling from 4.33 mg. % to 2.48 mg. %.

Thus, it seemed important to determine if priming doses are really necessary if constant plasma levels are desired. The first step in approaching this problem was to derive a theory which would have general application to the relationships between a constant rate of infusion (IV), the apparent volume of distribution (Vc), the renal clearance (C) and the plasma concentration (P) of a substance such as PAH or PACA.

These symbols are used in the following derivation:

(IV) = mg./min. infused at a constant rate
C = cc./min. of plasma cleared
Vc = cc. of body fluid in which substance is apparently dissolved
(UV) = mg./min. excreted
P = mg./cc. concentration in plasma or in Vc
M = total mg. of PAH in body at any time
t = time after beginning of constant infusion

The assumptions are made that loss of this substance from the body by routes other than the kidney is negligible compared to the rate of excretion by the kidney, and that the apparent volume of fluid in which it is dissolved remains constant.

The change in M with time will be the result of the sum of the rate of infusion and the rate of excretion:

$$(1)\ \frac{dM}{dt} = (IV) - (UV)$$

The plasma concentration at any time will be:

$$(2)\ P = \frac{M}{Vc}$$

$$(3)\ \text{thus } M = PVc$$

Substituting PVc for M in equation (1):

$$(4)\ \frac{dPVc}{dt} = (IV) - (UV)$$

Since Vc is constant we may rearrange as follows:

$$(5)\ \frac{dP}{dt} = \frac{(IV)}{Vc} - \frac{UV}{Vc}$$

Since by definition $\frac{(UV)}{P} = C$ or $(UV) = CP$

$$(6)\ \frac{dP}{dt} = \frac{(IV)}{Vc} - \frac{C}{Vc}P$$

Finally rearranging before integration,

$$(7)\ dp - \frac{(IV)}{Vc}\,dt + \frac{C}{Vc}\,Pdt = 0$$

Integration leads to the final equation:

$$(8)\ P = \frac{(IV)}{C}\left[1 - e^{-(c/Vc)\,t}\right]$$

From inspection of this equation, it is apparent that $\frac{(IV)}{C}$ is the plasma concentration which is approached with time and that the plasma concentration attained at any given time will be directly proportional to the rate of infusion (clearance and Vc remaining constant). This is illustrated in Figure 13.

The shape of the plasma concentration curves with changes in Vc or in clearance (C) are shown in Figure 14 and Figure 15 respectively. For these illustrations, values for clearance and rate of infusion were chosen which are in the range encountered in the determination of clearance by the constant infusion technique. The illustrations are theoretical curves derived by substitution of arbitrary reasonable values for (IV), (C) and (Vc) in equation (8).

In the relationships shown in equation (7), the only factor which is not directly determinable experimentally is the apparent volume of distribution (Vc). This can be calculated from experimental data. In Figure 16 there are two experimental curves of plasma level of PAH

after starting a constant infusion. The apparent volume of distribution of PAH was 4000 to 5000 cc. in these subjects.

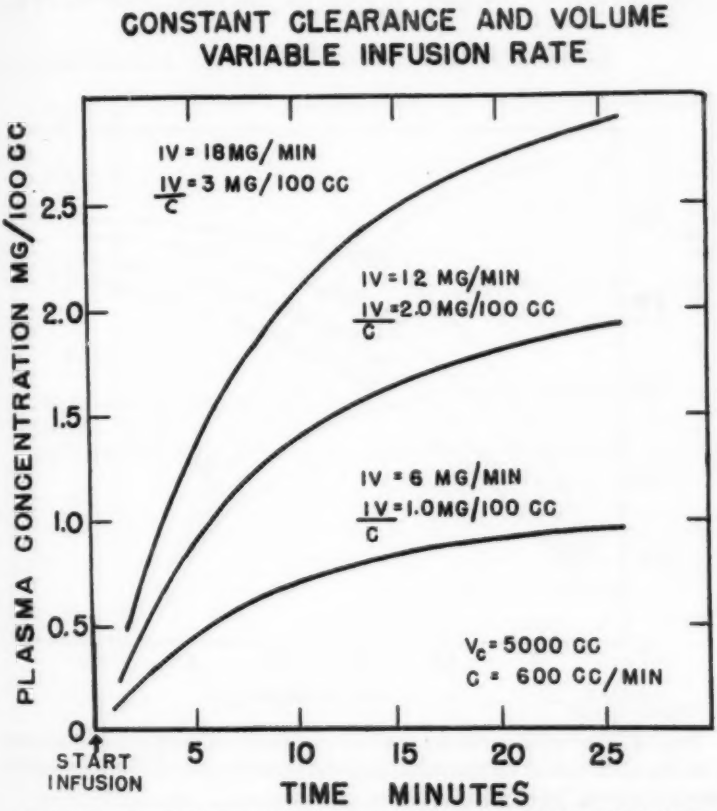

FIG. 13. Theoretical curves for plasma concentration of PAH or PACA derived from Equation 8 using various infusion rates with a constant clearance and constant apparent volume of distribution.

Thus, the plasma concentration, after beginning a constant infusion of PAH, follows the curve which we would expect if it were distributed in approximately five liters of body water. From the practical standpoint, both the theoretical analysis (Equation 8) and the experimental

facts serve to show that 20–30 minutes after starting a constant infusion the plasma level will have reached a maximum and constant value, and the time necessary to reach the maximum value will be independent of the value for rate of infusion and clearance, provided they are constant.

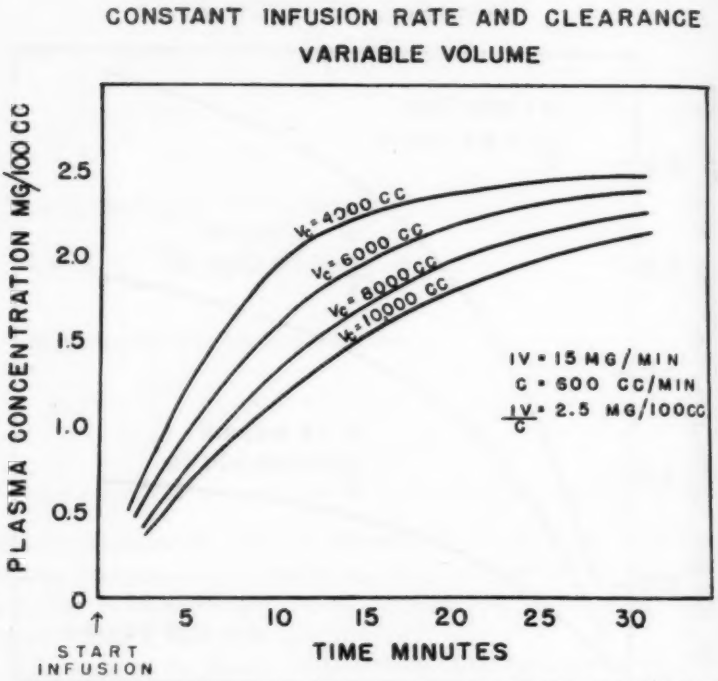

FIG. 14. Theoretical curves for plasma concentration of PAH or PACA derived from Equation 8 using various values for the apparent volume of distribution with constant rate of infusion and constant clearance.

Thus, the practice of giving "priming" doses of a substance such as PAH to be followed by a constant maintenance infusion is unnecessary. Since it usually requires 20–30 minutes for equilibration of substance such as PAH after the initial flooding of the circulation with the prim-

ing dose, the constant infusion by itself will accomplish the same constant blood level in the same time. Furthermore, there are other

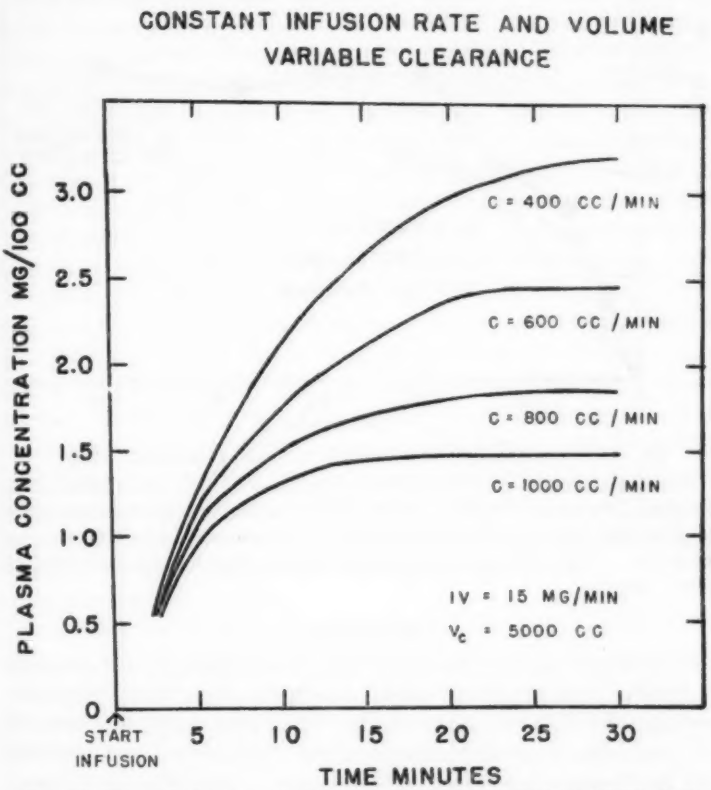

FIG. 15. Theoretical curves of plasma concentration of PAH or PACA derived from Equation 8 calculated using various clearance values with a constant rate of infusion and constant apparent volume of distribution.

objectionable features to priming doses. Side reactions such as flushing, chilliness and sweating usually occur during the injection of the initial concentrated solution used for a priming dose. These can be avoided by omitting the priming dose entirely in experiments during which a constant plasma level of PAH or of PACA is desired.

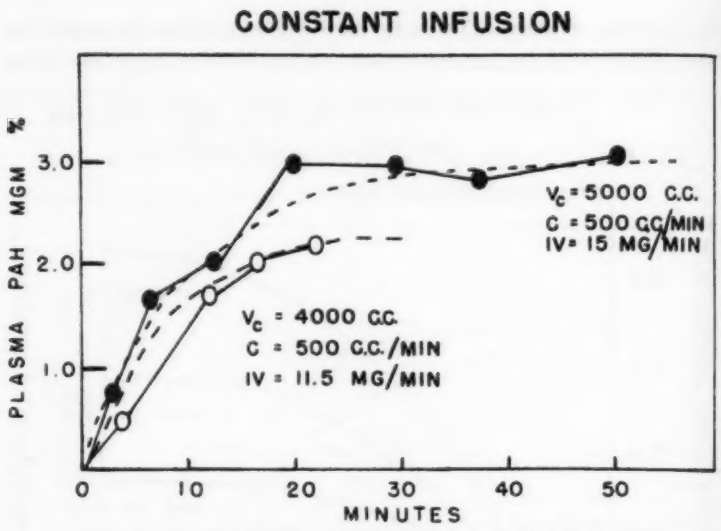

FIG. 16. CONSTANT INFUSION OF PAH WITHOUT A "PRIMING" DOSE

The dotted curves are obtained from Equation 8. By substitution of the experimental values for C (IV) and P, the values for Vc were obtained, giving the curves which best fit the experimental data. The constant infusion was started at time = 0. The solid lines are the plasma concentrations found in the two experimental subjects.

DISCUSSION

In the earlier studies with phenol red on the relationship of clearance to plasma concentration, single injections were used. Shannon, working with dogs (6), and Goldring, Clark and Smith with humans (7), gave single small and large injections of phenol red and concluded that the clearance was completely reversible. The clearance returned to the same high level with any dose used.

However, phenol red clearance was soon abandoned in favor of diodrast clearance as a measure of renal plasma flow, since diodrast yielded a higher clearance. The relationship of diodrast clearance to plasma concentration in man was described by Smith et al. (3), and this description is the basis of the relationship given in Figure 1. It appears, however, that no experimental facts were recorded showing that there is a constant maximal clearance range in man after single injections of

diodrast, or that self-depression of diodrast clearance is completely reversible.

Data on man after single injections of diodrast were recorded by White, Findley and Edwards (8). No comment was made on the fact which is apparent from their data that the clearance of diodrast showed a tendency to fall throughout the range of plasma concentration where the clearance was thought to be maximal and constant. Also, the clearances obtained with constant infusion at low plasma concentrations seem to be higher than the clearances after single injections.

The reason for the falling clearance and lack of reversibility of diodrast clearance after self-depression is not clear. Factors such as delay time, differences in arterial and peripheral venous plasma concentration, and changing urine flow would be unlikely causes of such large errors in the clearance determination. The facts that phenol red and PACA in man, and phenol red, PAH and PACA in the dog, yield reversible clearances and a constant maximal range after single injections make it likely that our findings are peculiar to diodrast and the human kidney, rather than to secondary circulatory or timing relationships. It is possible that diodrast is altered or conjugated in the body in a manner such that it would not be detected by the methods of determinations, which depend solely on the amount of iodine excreted. Whether different products of diodrast appear in human urine is now being investigated.

In the first studies on the excretion of hippuric acid derivatives by Smith (9, 10), it was reported that PAH in human urine was partly conjugated and that the clearances of PAH and PACA were equal both in dog and in man. Our finding, that there were increasing amounts of conjugated PAH in the urine with a falling plasma level, led us to suspect that either the excretion of the conjugated product was competing with PAH or that PAH was being conjugated by the kidney. The presence of a higher proportion of conjugated PAH in the urine than is present in plasma suggests that either the excretion of conjugated PAH takes precedence over the excretion of PAH or that the PAH is conjugated by the kidney.

It would be of great interest to determine the amount of PAH and "PACA" present in arterial and renal venous plasma simultaneously with the amount in urine. Demonstration of complete extraction of

PAH alone in renal vein plasma does not reveal whether there is any PACA present or if all PAH is present in the urine unmodified. Certainly it would be more desirable to use a substance which is not modified in the body and avoid the possibilities of interfering secondary substances.

Our findings emphasize that careful evaluation of many factors in the technique must be made in any experiment. With a perfectly constant plasma level, the proportion of conjugated PAH excreted or formed in the kidney may be small and constant. However, a change in plasma concentration may result in a change in the proportion of conjugated PAH in the urine and a change in clearance which is not a change in renal plasma flow. It should be emphasized again that there appears to be no definite data as to what chemical forms of diodrast and PAH appear in human urine after intravenous injection. When one instantaneous plasma determination is applied to the calculation of a clearance period lasting many minutes, absolute constancy of plasma concentration is essential for accuracy. The maintenance of a perfectly constant infusion is therefore necessary.

With a falling plasma concentration after a single injection, one eliminates the difficulty of maintaining a perfectly constant infusion, and one can describe the course of the plasma level accurately since it follows a consistent curve. However, other difficulties are introduced by using a falling concentration. One must limit the length of the observations so that the plasma concentration remains within the range of maximal clearance. Also, there is the possibility of arteriovenous differences in plasma concentration. Such differences would be more likely to cause error when the plasma concentration has fallen to low levels, if the arterial is not proportional to the venous plasma concentration.

Since it is desirable to determine clearances on the same plasma which reaches the kidney, namely arterial plasma, we are now investigating a method for analysis of "arterial" plasma obtained from finger blood. Enough plasma (0.2 cc.) can easily be obtained from blood from a finger tip puncture. It may be possible then to determine clearances after a single injection of PACA, eliminating any error due to arteriovenous differences in plasma concentration and obtaining

more frequent samples without venapunctures and loss of greater quantities of blood.

SUMMARY

1. Constant plasma clearances were obtained with falling plasma concentrations after single injections of PAH and PACA in dogs and with PACA in man.

2. Falling clearances were obtained in man, with falling plasma concentrations of diodrast and PAH after single injections, and with plasma concentrations sustained by infusions after temporary elevation of plasma level.

3. Conjugated PAH appears in increasing proportion to PAH in the urine with falling plasma concentration, and the clearance of conjugated PAH becomes higher than the clearance of PAH.

4. The many factors which could produce invalid clearance measurements of renal plasma flow are discussed. Two factors may, under some circumstances, be important: modification of the injected substance; a difference between arterial and venous plasma concentration.

5. A general theory is derived of the relationship between rate of infusion, plasma concentration, and plasma clearance of a substance excreted at a rate proportional to the plasma concentration.

6. If a constant plasma concentration is desired, a "priming" dose before the sustaining infusion is unnecessary.

7. A method of preparation and determination of PACA is described.

Dr. E. K. Marshall and Dr. K. Blanchard of the Department of Pharmacology have given valuable advice and criticism.

We are indebted to Dr. Margaret Merrell, Associate Professor of Biostatistics, School of Hygiene and Public Health, Johns Hopkins University, for aid with the theoretical analysis.

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AN UNUSUAL CASE OF SQUAMO-BASAL CELL CARCINOMA

TORRENCE P. B. PAYNE

From the Department of Pathology, The Johns Hopkins University Medical School Baltimore, Maryland

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It is well known that basal cell carcinoma is not a morphologically closed entity; there are transitional forms between the histologically pure basal cell carcinoma and the prickle or squamous cell carcinoma (1). Basal cell tumors in which there is some differentiation towards squamous cell carcinoma display a greater degree of malignancy than those in which no such differentiation is present (2, 3). Moreover, these transitional forms, or squamo-basal cell carcinomas, in the majority of cases are relatively resistant to roentgen-ray and radium treatments as compared with pure basal cell tumors (3). The purpose of the present report is to describe a squamo-basal cell carcinoma of extreme malignancy and extraordinary morphology.

A sixty eight year old white man was admitted to the private surgical service of The Johns Hopkins Hospital (History No. 255034) on June 14, 1946, complaining of intermittent episodes of left upper quadrant abdominal pain of two months duration.

There had been several previous admissions to the hospital—in June, 1923 for tonsillectomy and adenoidectomy, and in March, 1942 for appendectomy. Of greater interest, however, was a series of admissions that started on February 10th, 1941 when a chalazion was removed from the inner third of the left upper eyelid at the Union Memorial Hospital. On microscopic examination it was diagnosed: "Basal Cell Epithelioma." On April 29th, 1943 at the Johns Hopkins Hospital a small nodule was removed from the left upper eyelid at the same site as the chalazion excised in February, 1941. On microscopic examination it was diagnosed: "Squamous Cell Carcinoma." On October 1st, 1943 at the Union Memorial Hospital almost the entire inner half of the left upper eyelid was removed with a portion of tarsal cartilage. No pathological report on this tissue is available. A plastic repair on the eyelid was done. On April 6th, 1944 at the Union Memorial Hospital a small nodule at the site of the scar of the last operation was excised together with a swollen preauricular lymph node. Both specimens were reported as showing a similar type of tumor and the diagnosis was: "Carcinoma, skin appendage, upper eyelid." On September 17th, 1944 at the same hospital a growth was removed from the outer half of the left upper eyelid and a swollen lymph node was excised

from the left side of the neck. The pathological report was: "Carcinoma, epidermoid, basal cell, with metastasis to lymph nodes."

During the period from July, 1941 until April, 1946 the patient received large amounts of roentgen and radium treatment directed first to the lesion on the left upper eyelid and later to the left neck and sub-maxillary region. Under this therapy he developed hypothyroidism in early 1945 so that thyroid extract, one grain per day, had to be administered.

The family history was entirely non-contributory. The personal history revealed a life long tendency for eczematoid eruptions to occur particularly on the skin of the face.

The present illness began in April, 1946 when there occurred episodes of left upper quadrant abdominal pain, bearing no relationship to meals or any other recognizable factor. At about the same time he began to suffer from constipation. These symptoms became worse and anorexia developed. Then he noticed progressive and severe limitation of stomach capacity, liquid foods being tolerated more easily than solid. There was one episode of vomiting and several periods of severe nausea. The patient had become weaker and had lost about ten pounds in weight in the two months prior to admission.

On admission to hospital the temperature was 97.6°, the pulse 80, respirations 20 and blood pressure 100/60. The patient was an elderly male in no apparent distress. His skin showed evidence of recent weight loss. The left upper eyelid showed the result of excision of portions of it and subsequent graftings. No nodules were felt in it. Under the angle of the left jaw there was a hard mass (size not mentioned) which felt like a lymph gland, discrete but attached to the deep tissues. The skin was indurated over the left posterior cervical region. The thyroid could not be palpated. There was no generalized lymph node enlargement, but there were several subcutaneous nodules, each measuring roughly about 7 mm. in diameter, over the thorax and abdomen. The lungs were clear on percussion and auscultation. The heart was normal. The abdomen was relaxed and there was no tenderness or muscle spasm. The liver was palpable two fingers breadth below the costal margin. The spleen could not be felt. In the epigastrium there was resistance, but no well defined mass could be felt. Rectal examination revealed a diffusely enlarged and moderately firm prostate. Neurological examination was negative.

Examination of the blood showed Hbg. 11 gm, WBC 6,200. The urine was negative. The blood serological test for syphilis was negative. No organic lesion could be demonstrated by a barium enema. A gastrointestinal series was done; the lesser curvature of the stomach was very rigid and no peristaltic waves were seen going through that portion of the stomach. This was interpreted as suggesting the presence of an infiltrative lesion in that region.

The clinical diagnosis was carcinoma of the stomach.

On June 17th, 1946 an exploratory laparotomy was performed. The stomach was described as being a solid mass of tumor and operative removal of its was con-

sidered impossible. A biopsy of an enlarged omental lymph node was made. The pathological diagnosis was: "Squamo-basal cell carcinoma."

Postoperatively the patient's condition gradually became worse and he died on August 1st, 1946.

The autopsy (No. 20074) was performed 12 hours after death. The report is abbreviated, but no pertinent observations are omitted.

The body is that of a well developed, poorly nourished, white adult male, measuring 169 cm. in length and weighing 59.5 kg. The superficial lymph nodes are not palpable except in the cervical and axillary regions; in the latter location a firm nodule about 2 cm. in diameter is felt. On the left upper eyelid there is a small scar causing the eyelid margin to be irregular. No skin lesions are described.

There is about 1500 cc. of clear fluid in the peritoneal cavity. The peritoneal surfaces are everywhere studded with small yellowish-white tumor nodules each measuring about 5 mm. in diameter. These nodules extend in linear fashion along the mesenteric attachment of the entire small intestine. In the subdiaphragmatic and pelvic regions they are more numerous and larger in size. In the left hypochondrium the stomach and transverse colon are bound together by tumor tissue. The peripancreatic and para-aortic lymph nodes are all enlarged and firm; about the origins of the renal arteries there are lymph nodes measuring as much as 2 cm. in diameter and having bulging granular grayish-white cut surfaces.

Each pleural cavity contains about 500 cc. of clear fluid. There are no adhesions. The pericardial cavity is normal. The site of the thymus is occupied by fatty tissue; there is no mass in this region. The mediastinal lymph nodes are enlarged, especially at the bifurcation of the trachea where several of them, each measuring about 1 cm. in diameter, are seen; they are firm in consistency and on section present a bulging grayish white cut surface with small areas of anthracotic pigmentation.

The heart weighs 350 grams. The valves are normal. The orifices of both coronary arteries are narrowed and there is sclerosis and calcification of all the coronary arteries with marked eccentric narrowing of their lumina. Two tumor nodules are seen in the myocardium; one of these measures about 3 mm. in diameter and lies in the anterior wall of the left ventricle towards its base, the other forms a greyish pearly area about 1 cm. in diameter high up in the interventricular septum.

The pleural surfaces of both lungs are smooth. The lungs are air-containing. The bronchial mucosa is normal everywhere.

The liver is firmly adherent to the underlying stomach. The cut surface shows slight central atrophy and congestion and a single tumor nodule about 1 cm. in diameter is seen. The gall bladder is thin walled and contains numerous small facetted stones.

The spleen weighs 100 grams. The capsule is thickened in one area.

The oesophagus is normal. The stomach is adherent to the liver, pancreas and a portion of the transverse colon. Its mucosa is flat and there is no ulceration. The stomach wall is infiltrated with greyish white tumor tissue especially in the region of the fundus where the wall measures about 1 cm. thick. The duodenum

is normal. The mucosa of the jejunum and upper ileum is normal. In the lower ileum two or three greyish white tumor nodules are seen projecting from the mucosal surface; the mucosa over these areas is ulcerated so as to form a rounded crater in each nodule. The mucosa of the large intestine is normal. The mesenteric lymph nodes are not enlarged.

The peripancreatic lymph nodes are replaced by tumor and many tumor nodules are seen in the pancreas itself.

The medulla of the left adrenal is replaced by a greyish white nodule of tumor tissue measuring about 1 cm. in diameter. In the medulla of the right adrenal a smaller nodule measuring about 5 mm. in diameter is seen.

The kidneys are normal except that the pelvis and calyces of the left kidney are slightly dilated. The right ureter is delicate and patent; the left ureter and the inferior portion of the left renal pelvis show thickening of their walls by invading tumor tissue—the left ureteral wall throughout its length measures about 5 mm. in thickness. The ureteral openings into the bladder are patent. The prostate is slightly enlarged and on section several small firm nodules are seen in its right lobe.

The thyroid is very small and difficult to identify grossly. The paratracheal lymph nodes are slightly enlarged and are invaded by tumor. The tongue, pharynx, larynx, and trachea are normal. The tonsils are not present.

The bone marrow of the ribs appears normal. In the lumbar vertebral marrow there are pale yellowish areas suggesting tumor metastases.

The brain and hypophysis are grossly normal.

Study of the microscopical sections from the organs show little apart from invasion by tumor. There is atrophy and scarring of the thyroid gland, probably caused by irradiation. The pancreas shows dilated ducts, atrophy and scarring due to obstruction of its duct system by tumor. In the anterior lobe of the hypophysis there is a poorly preserved adenoma. There is lobular pneumonia in the lungs and a small adenoma is seen in the prostate.

The tumor invades the following tissues: cervical, mediastinal and para-aortic lymph nodes, peritoneum, stomach, liver, pancreas, capsule of spleen, adrenals, lung, left ureter and renal pelvis, lower ileum, myocardium and vertebral bone marrow.

The remarkable feature of this tumor is that it is composed, in places, of round cells with deeply staining nuclei and scanty cytoplasm. These cells grow in the most diffuse manner, suggesting the appearance of a lymphosarcoma (Fig. 1). In all of the sections there are tiny round areas of squamous cells with keratinization and pearl formation. In some places these areas lie in the midst of the diffuse growth of round cells so that the tissue bears a striking resemblance to thymic tissue, the tiny, round squamous foci suggesting Hassall's bodies (Fig. 2). In other places the pearls are surrounded by spindle shaped cells with hyperchromatic nuclei and scanty cytoplasm. These cells tend to

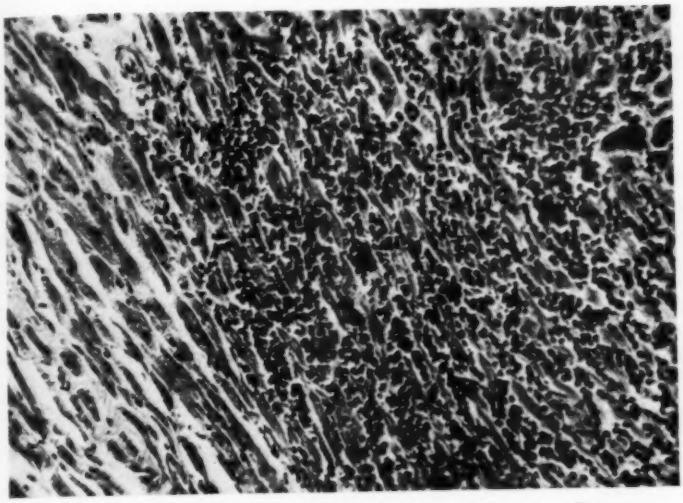

FIG. 1. TUMOR IN MYOCARDIUM. NOTE TYPE OF CELL AND DIFFUSE INFILTRATION SUGGESTING LYMPHOSARCOMA

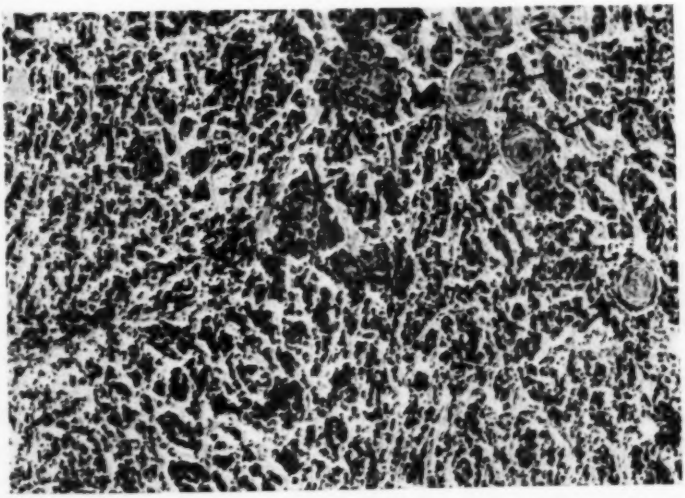

FIG. 2. TUMOR IN CONNECTIVE TISSUE AROUND RENAL ARTERY. DIFFUSE GROWTH OF ROUND CELLS WITH SQUAMOUS FOCI (ARROWS) SUGGESTING THYMIC TISSUE

cling together rather than to be diffusely scattered. There is no peripheral palisading of cells around these groups. It is in these latter areas that the similarity to squamo-basal cell carcinoma is evident. No intercellular bridges are present between any of the cells.

Review of the series of biopsies taken from the left upper eyelid and involved lymph nodes during the patient's life makes it reasonably clear that the tumor originally arose from the epithelium of the face and later metastased widely. The first section removed from the eyelid over five years before death shows a squamo-basal cell carcinoma in which the basal elements are closely grouped together and surrounded by a definite palisade of cells. The subsequent biopsies show a gradual transformation into a tumor in which the cells, for the most part, no longer form bands or coherent masses but infiltrate not infrequently as separate cells (Fig. 1) which have round or elongated hyperchromatic nuclei and very little cytoplasm.

DISCUSSION

This tumor ran a much more malignant course than is seen in basal cell tumors, and displayed a marked insensitiveness to irradiation.

Microscopically, although the tumor seemed to fall into the group of squamo-basal cell carcinomas, its structure was decidedly unusual. The cells, for the most part, did not cling together to form nests and strands and distinct groups as in the usual basal cell carcinoma. On the contrary they tended to infiltrate diffusely, often as single cells, and indeed in many microscopic fields the tumor could easily be mistaken for a lymphosarcoma (Fig. 1) or, in areas where there were foci of squamous cells with keratinization, for thymic tissue (Fig. 2).

As far as can be determined, no similar case has been reported. However, in his description of "transitional cell carcinomas", Ewing (4) makes the statement: "In very active cases of this type, the cells may lose most or all of their epithelial characters, and the tumor may grow diffusely and be difficult to distinguish from lymphosarcoma or lymphoepithelioma." But these carcinomas generally fail to show squamous cells (4).

SUMMARY

This report describes a case of extremely malignant squamo-basal cell carcinoma, which was remarkably peculiar in that the basal cell

elements, in many places, grew in a diffusely invading manner suggesting the morphology of a lymphosarcoma. Associated with the basal cells, there were minute, globoid squamous cell formations.

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GAUCHER'S DISEASE WITH THROMBOCYTOPENIA, AN INSTANCE OF SELECTIVE HYPERSPLENISM

F. W. DAVIS,* ABRAHAM GENECIN,† AND ERNEST W. SMITH‡

(From the Medical Clinic, The Johns Hopkins Hospital)

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A Case Report

In recent years great interest has been manifested in the concept of hypersplenism, a term denoting reduction of the formed elements of the peripheral blood in association with splenomegaly. This effect has been attributed by some to altered phagocytic function of the reticulo-endothelial cells of the spleen, and by others to the influence of a possible humoral agent of splenic origin on the bone marrow when splenic hypertrophy occurs. Hypersplenism may be selective or panhematocytopenic, i.e., depression of any one of the formed blood constituents or any combination of these elements may be observed. Recently a case was observed by us in which selective hypersplenism (thrombocytopenia) was associated with the splenomegaly of Gaucher's disease.

CASE REPORT

S. D., a 32 year old Alabama housewife, was admitted to the Johns Hopkins Hospital 7/25/47, on the service of Dr. J. E. Howard, complaining of a mass in the abdomen which had been known to be present for 2½ years. The family and past histories yielded no material pertinent to the present illness. A tendency to bruise and bleed easily, the latter having been noted particularly following dental extractions, had been manifested for many years. Three years prior to admission, a prolonged hemorrhage occurred following a miscarriage. One year later, following delivery of a normal child, there occurred a hemorrhage of sufficient severity to require a transfusion. During this pregnancy, 2½ years prior to admission an enlarged spleen was discovered. It was the patient's opinion that the mass had not increased in size since that time. The patient had always been of tense, anxious disposition and had not changed in this respect over a period of ten years. In view of the tendency to bleed and splenomegaly, the patient was referred for study.

Physical examination revealed a small, poorly nourished young woman of

* Assistant Resident.

† Resident.

‡ Interne.

swarthy complexion. There were firm, discrete small lymph nodes palpable in the left axillary and left inguinal regions. The skin was warm, moist, with marked palmar and plantar hyperhydrosis, and moderate dermatographia was demonstrable. Bilateral pingueculae were present. There was slight bleeding from the gums. A massive spleen was felt, which was firm, non-tender, extended to the pelvic brim and occupied almost the entire left abdomen. The physical examination was otherwise not remarkable.

Laboratory findings of significance included: normal urinalysis and stool examination; doubtful serologic test for syphilis;[1] normal RBC (4.71 million), with hemoglobin of 13.5 Gm., hematocrit 41.8 (MCV—89, MCH—29, MCC—32); normal WBC (6,500), with Juvenile neutrophiles 7.0%, Segmented neutrophiles 65.0%, Eosinophiles 1.0%, Atypical lymphocytes 0.5%, lymphocytes 21.5%, monocytes 5.0%; normal osmotic fragility test; platelet count by the Rees-Ecker method 62,000; markedly positive tourniquet test with showers of petechiae appearing after five minutes of compression with pressure midway between systolic and diastolic tensions (115 mm. of Mercury); Bilirubin 1.6 mgm. per 100 cc. of serum of which less than 0.8 mgm. reacted directly; cholesterol 110 mgm. per 100 cc. of serum; cephalin flocculation 3+, thymol turbidity 11.2 units.

In view of the splenomegaly and thrombocytopenia, a sternal puncture was performed; Dr. C. Lockard Conley interpreted this as follows: "There is an abundance of rounded and ovoid cells 30 to 50 MU in diameter, of somewhat indistinct outline, the cytoplasm of which stains poorly, with faint bluish cast and a fine wavy fibrillar structure. The nuclei are small and distinct, often eccentrically placed and many cells are multinucleated." These cells were felt to be typical Gaucher's cells. The megakaryocytes were numerous, but the platelets were reduced in quantity.

After establishment of the diagnosis of Gaucher's disease, it was felt that the thrombocytopenia and megakaryocytic arrest were on the basis of secondary hypersplenism, and in view of the history of bleeding it was deemed advisable to perform a splenectomy. On 8/2/47 a splenectomy and liver biopsy were carried out by Dr. Alfred Blalock under nitrous oxide, ether, and oxygen anaesthesia, with removal of a spleen weighing 2300 gms. Within four hours postoperative, the platelets had increased to 116,000 and by the second postoperative day had reached 241,000. On the eleventh postoperative day the platelets reached 424,000. Concomitant with the rise in platelets, a rise in leucocytes to 19,500 on the second and 14,000 on the eleventh postoperative days and a rise in hematocrit to 45 were observed. No significant change in differential count or cell indices was observed. Postoperatively the tourniquet test was entirely negative on the third day, the cephalin flocculation fell to 2+, the thymol turbidity to 8.8 units, the cholesterol rose to 181 mgm per 100 cc. of serum, and the Bilirubin fell to less than 0.8 mgm per 100 cc. of serum on the eleventh day. The prothrombin concentrations both pre- and postoperatively ranged between 50 and 75% of normal.

[1] Slide test persistently positive, Wassermann negative. It was our opinion that this was a biological false positive.

Microscopic examination of the liver biopsy specimen showed normal liver cells but many large Gaucher's cells in the liver capillaries. The spleen was infiltrated with these cells which formed nests and strands throughout the entire splenic pulp so that little remained of either the red or the white pulp; there were only a few generally atrophic Malpighian bodies scattered throughout the spleen (Dr. Richard Follis, Jr.).

The postoperative course was entirely uneventful in all other respects and patient was discharged, greatly improved, with the diagnosis of Gaucher's disease and secondary thrombocytopenia.

Twelve months after operation the platelet count remained above preoperative levels, 192,500, the hemoglobin was 70%, the WBC 18,450 with a normal differential count, and there had been no further tendency to bleed or bruise. (Courtesy of Dr. W. L. Miller, Gadsden, Ala.)

DISCUSSION

We believe that this case illustrates selective depression of a circulating blood element, in which dramatic remission followed removal of the spleen laden with Gaucher's cells, and that the thrombocytopenia was a manifestation of hypersplenism. Such diseases as congenital hemolytic icterus, idiopathic thrombocytopenic purpura, and primary splenic neutropenia have been considered varieties of hypersplenism. However, the picture of hypersplenism has been reported in association with many other diseases in which splenic involvement occurs, including Hodgkin's disease, tuberculosis, sarcoidosis, Banti's syndrome, etc. (1–7).

It is well known that reduction of platelets is a prominent feature of Gaucher's disease, and purpuric manifestations are among the most common physical findings (8–12). In the great majority of such instances the thrombocytopenia is a manifestation of more general reduction of all of the circulating blood elements, with anemia and neutropenia likewise present. In his series of 220 splenectomies, Doan (3) found only two instances of Gaucher's disease, in both of which panhematocytopenia was present, and both were relieved by splenectomy. Other observers have reported similar improvement in the hematologic picture following splenectomy in Gaucher's disease (3–10). That the thrombocytopenia is due to megakaryocytic arrest and not to myelopthisis has been recently emphasized by Dameshek and Miller (6) and confirmed by our case.

The patient herein described demonstrated as the only hematologic abnormality a marked thrombocytopenia with megakaryocytic arrest.

The diagnosis was established by the finding of typical Gaucher's cells in the material obtained by sternal puncture, and following splenectomy the platelet count rose to normal levels. This case, therefore, appears to be one of selective hypersplenism secondary to the splenomegaly of Gaucher's disease.

SUMMARY

An instance of thrombocytopenia without evidence of neutropenia or anemia on the basis of hypersplenism secondary to Gaucher's disease is reported. Splenectomy was followed by return of the hematologic picture to normal.

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PROCEEDINGS OF THE MEETING OF THE JOHNS HOPKINS MEDICAL SOCIETY

Held in Hurd Hall, November 8, 1948

Dr. Follis: I should like to open this first meeting of this year of The Johns Hopkins Medical Society.

The first speaker on the program this evening is Dr. Charles G. Zubrod, of the Department of Pharmacology and Experimental Therapeutics and Medicine. The title of Dr. Zubrod's paper is "The Need for Intrathecal Therapy in the Treatment of Experimental Meningitis".

The Necessity of Intrathecal Drug in the Treatment of Experimental Meningitis. Gordon Zubrod. (Department of Pharmacology and Experimental Therapeutics, The Johns Hopkins University.)

An experimental meningitis has been induced in cats by intracisternal injection of K. pneumoniae. This infection is uniformly fatal in untreated animals. Treatment with intramuscular streptomycin produces a significant number of cures only when relatively large doses are given. Treatment with intrathecal streptomycin or with simultaneous intramuscular and intrathecal drug indicates that cure is a function of the amount of drug in the cerebrospinal fluid. For optimum results in the treatment of this experimental meningitis, one must effect the presence of streptomycin in the cerebrospinal fluid. This can be accomplished certainly by intracisternal administration, and probably by the intramuscular route if large enough doses are given. The clinical implications of these data are discussed.

Dr. Follis: Dr. Zubrod's paper is open for discussion.

Dr. Marshall: I have been very much interested in this question since the sulfonamides came out several years ago. Whether it is necessary to give both intrathecal and parenteral administration of therapeutic agents for meningitis has been discussed frequently, but as far as I know, this study of Dr. Zubrod's is the first attempt to answer the question experimentally. Personally, I have always thought it was unnecessary to give the drug intrathecally. Apparently, I have been wrong. It appears that drug in spinal fluid alone is effective, whether it gets there by being put in intrathecally or by passing the blood-brain barrier.

I think Dr. Zubrod emphasized, and I would like to emphasize again, that with very large doses one obtains as good results with intramuscular doses alone as with the combined dosage. I think this is a very important point and must be kept in mind.

Dr. Dowling—I happen to have a copy of his paper; it is not published yet, but is in the press—actually had 10 cases of pneumococcic meningitis that were treated with intramuscular penicillin alone; of these, 9 recovered. They received penicillin

intramuscularly, 12 million units per day, getting it every 2 hours. The very large doses given, allowed a sufficient amount of penicillin to pass over into the spinal fluid to accomplish the result.

Dr. Follis: Are there any other comments on Dr. Zubrod's paper?

Dr. Rich: Gentlemen, I was a little baffled—perhaps I understood it wrongly—but I thought Dr. Zubrod suggested that you should give large intramuscular doses together with the intraspinal. As I see it from the figures, it seems to me it doesn't matter what you give intramuscularly if you give intrathecal doses.

Dr. Zubrod: The reason I suggested that, Dr. Rich, was because in the treatment of clinical meningitis the antibiotics are usually given in the lumbar subarachnoid space. The distribution of drugs from the lumbar region upwards is sometimes erratic and rather limited. If you are using small doses of intrathecal drug in the lumbar area, I think you should also use enormous doses intramuscularly in the hope that the blood-brain barrier will allow passage of the drug into the ventricular and cisternal fluid. The results with the experimental meningitis were accomplished with intracisternal injections; one cannot be sure that the same results could be achieved by the use of the lumbar route.

Dr. Follis: Is there any other discussion of Dr. Zubrod's paper?

The second presentation this evening will be by Dr. Allan L. Grafflin of the Department of Anatomy, whose title is "The Excretion of Fluorescein by the Liver under Normal and Abnormal Conditions".

Studies of Fluorescein Excretion by the Liver Under Normal and Abnormal Conditions. ALLAN L. GRAFFLIN. (Department of Anatomy, Johns Hopkins Medical School.)

The excretion of fluorescein by the liver, under normal and abnormal conditions, has been investigated in male specimens of Rana pipiens, receiving fluorescein (uranin) in standard dosage of 30 mg. per kg. The course of excretion was observed in vivo with the fluorescence microscope.

Under normal conditions, after injection of fluorescein the dye appears promptly in the plasma, then in the hepatic cells and the bile canaliculi. The peak of excretion (all bile canaliculi sharply delineated at high intensity, all hepatic cells diffusely stained at low intensity) is reached in approximately one hour at ca. 15 degrees C. The sequence of events from the peak of excretion to complete elimination from the liver is essentially a progressive diminution in fluorescent intensity of the dye to non-detectable levels, achieved first in the hepatic cells, then (much later) in the bile canaliculi. As long as the bile canaliculi are delineated they persist as a continuous system, without detectable interruptions in their course. Elimination is usually complete in 24 hours or less. Localized enlargements of the axial bile canaliculi and dilatations at the distal ends of their intercellular branches are repeatedly observed, and are regarded as normal variants of the bile canalicular picture during the excretion of fluorescein.

The present observations upon the normal mechanism of fluorescein excretion

have failed to confirm the findings of A. Hirt, J. Ansorge and H. Markstahler (Ztschr. f. Anat. u. Entwickl., 109, 1, 1939) in the later stages of excretion (extensive sprouting of fluorescein-containing vacuoles from bile canaliculi, widespread intracellular vacuole formation, discontinuity of bile canaliculi, "lakes" of fluorescein). However, the whole range of these findings has been observed in animals subjected to highly abnormal conditions; and the distortion of normal function when the liver is subjected to prolonged observation, in vivo, with the fluorescence microscope has been clearly demonstrated.

The author's observations have been reported in full elsewhere (American Journal of Anatomy, volume 81, page 63, 1947).

Dr. Follis: Dr. Grafflin's paper is open for discussion.

If there is no discussion we will pass on to the third and final paper of this evening's program, which is by Dr. Manfred M. Mayer of the Department of Bacteriology of the School of Hygiene, who will talk on "Recent Advances in Immunochemistry".

Recent Advances in Immunochemistry. MANFRED M. MAYER. (Dept. of Bacteriology, School of Hygiene and Public Health.)

Precise chemical methods for the quantitative measurement of the precipitin and agglutinin reactions were introduced about 15 to 20 years ago and have since proved invaluable to theoretical studies in immunology as well as to the solution of various problems in medicine and biology.

For example, accurate determinations of the ratio of albumin to globulin in serum and spinal fluid have been made by immunological technics, with results corresponding to those obtained by electrophoretic analysis. Similarly, it has been shown that type-specific pneumococcal polysaccharide can be used as an indicator substance in determinations of blood volume. In studies on plant and animal viruses antibody has been used as a specific reagent for quantitative determination of the degree of purification. Thus it could be shown that highly purified tobacco mosaic virus was free from constituents of the host plant, while highly purified influenza virus contained large amounts of host material. In studies on anaphylaxis and hypersensitivity application of the quantitative precipitin method has made it possible to secure more strictly comparable and reproducible results. Striking advances have also been made in blood group studies.

Subsequently, the quantitative precipitin method has been extended to the chemical determination of complement in guinea pig, human and bovine serum. These studies proved that complement is an actual substance, namely a protein constituent of serum, rather than a "colloidal state" of fresh serum, as had been claimed not so many years ago.

More recently, technics have been developed for the quantitative and objective measurement of the hemolytic activity of complement, and their use has made

possible a considerable improvement in the reliability of complement fixation tests. In the course of these studies it could be shown that Mg^{++} is an essential co-factor in hemolysis by antibody and complement, a fact of theoretical significance as well as practical utility.

During the past 2 years the work in our laboratory at the School of Hygiene has been concerned mainly with the mechanism of hemolysis by antibody and complement. Studies on the velocity of immune hemolysis have shown that in the presence of excess complement and limited amounts of antibody the kinetics of hemolysis resemble those of enzymatic processes. Assays of hemolytic antibody should therefore be based on velocity measurements rather than end point titrations. Such a technic has now been developed and is being utilized at present in studies on purified antigens from erythrocytes and on the nature of the various antibodies present in anti-erythrocyte sera. It is hoped that in the future similar technics can be used in the immunochemical analysis of antireticular cytotoxic serum which has been claimed to be beneficial in wound healing.

In general, immunochemical investigations in recent years have been directed toward the use of highly purified antigens and the quantitative chemical measurement of their interaction with antibody.

Dr. Follis: Dr. Mayer's paper is open for discussion.

Dr. Turner: Mr. Chairman. Immunology is frequently spoken of as the distinguished offspring of bacteriology. The two touchstones of immunology are the antigen, on the one hand, and the antibody on the other. An antigen is ordinarily defined as a substance that provokes the production of an antibody. And what is the definition of an antibody? A substance that reacts to an antigen!

So, however diffuse these definitions might be, it seems evident that the materials with which the modern immunologist works are far more precise. Dr. Mayer has indicated ways in which these methods might be used. I need only to point out that there are many practical applications—among others, in the further identification, purification and isolation of antigens used in vaccines. The typhoid bacillus, for example, is a most complex organism containing numerous antigens—flagella, somatic and the so-called V. I. antigens—yet despite this knowledge we simply grow the organism in bulk, add a little phenol or formalin, and inject this into human beings in the hope that we will get some immunity. I think it is obvious that with modern immunochemical methods a great deal of work can be done toward developing more potent vaccines, not only for typhoid, but for various other diseases.

Dr. Follis: Is there any other discussion of Dr. Mayer's paper?

Dr. Vandegrift: How do you calibrate your antigen. How do you stabilize your antibody?

Dr. Mayer: You mean the hemolytic antibody? We usually maintain hemolytic sera under aseptic conditions and, if possible, we keep the sera frozen. Under these conditions they are stable. That may not be true for all antibodies, but

certainly for those we work with. We check occasionally and we rarely run into difficulty, provided serum is not contaminated by micro-organisms. Does that answer your question?

Dr. Vandegrift: Not entirely. How do you freeze them? At what temperature are they maintained.

Dr. Mayer: We put them in the deep freeze at a temperature of about −20°C. We just like that because that means we can go back into a bottle occasionally without having to take extensive aseptic precautions, but I have also kept many sera in the icebox in the liquid state without loss of activity. We have made analyses of the weight of precipitable antibody, even years apart, and never found very much of a difference. I can't recall a single instance where a serum analyzed by the quantitative method has seriously deteriorated. Of course, I might add that we usually work with sera from hyper-immunized rabbits, and what I am saying need not be true for convalescent sera containing very low levels of antibody.

Dr. Follis: Is there any other discussion of Dr. Mayer's paper?

Dr. Rich: I would like to ask whether it is conceivable that the antibody is not acting as if it were an enzyme but is acting on the cell and the lysis of a cell releases it.

Dr. Mayer: Well, I'll tell you, Dr. Rich, we have looked for that and I didn't mention the experiments tonight, but we have made experiments that show such an effect What we did, essentially, was to lyse a suspension of red cells completely with antibody and complement and then collect the stromata, i.e. the insoluble structural parts of the red cell, by centrifugation. These were then washed very extensively—six, seven or eight times—until there was not the slightest trace of antibody in the washings, and then these stromata to which the bulk of the antibody is still bound were suspended in saline and mixed with a portion of red cells. No hemolytic antibody was added. On incubation nothing would happen. However, on addition of complement the new portion of cells lysed.

Now, that doesn't prove conclusively that antibody combines with a red cell, does its work, comes off again and goes on to another red cell, because it is just possible—in view of the fact that antibodies are presumably multivalent, that an antibody molecule attached to a stroma particle with one of its reactive groups can react with a red cell by means of another reactive group, and will thus accomplish lysis of the new red cell. That is a proposition that can be checked experimentally and it is just the pressure of other experiments that has kept me from investigating it. I think one can readily follow that microscopically.

I might add, of course, that I feel quite strongly that the experiments described do not prove that antibody acts like an enzyme, but they suggest that there is a similarity in behavior of hemolytic antibody and enzymes.

Dr. Rich: Of course, I meant it in a little more crude fashion than that. I'd like to know if it really does enter in the combination, erupt mechanically or is shaken loose.

Dr. Mayer: Well, one can show very readily that the bulk of the antibody that

you put into a red cell suspension with complement is still attached to the stromata after lysis. It is necessary to subject the suspension of stroma to special treatment in order to dissociate antibody. The results we obtained are probably due to the fact that a very small amount of antibody is in the fluid phase in equilibrium with antibody in combination. When red cells are added to the stromata combined with antibody, the equilibrium is displaced and antibody transfers from the stromata to the red cells.

Dr. Follis: Is there any further discussion?

The meeting is adjourned.

BOOK REVIEWS

(These reviews represent the individual opinions of the reviewers and not necessarily those of the members of the Editorial Board of this Journal)

Handbook of Communicable Diseases, 2nd edition. By FRANKLIN H. TOP. 992 pp. Illus. The C. V. Mosby Company, St. Louis. $9.50.

In the composition of the 1947 edition of his Handbook of Communicable Diseases Dr. Top has encountered the difficulties of keeping abreast of the rapidly changing fields of new chemotherapeutic, antibiotic, and antihistaminic agents. No sooner is a book such as this on sale, and perhaps before its final proofreading than there are major changes that might well be made. Such situations may be considered as beyond the author's control. This new edition is a distinct improvement over its 1941 parent. The addition of new chapters such as those by T. J. Abernathy on Primary Atypical Pneumonia and T. Francis Jr. on Infectious Hepatitis are worthwhile as concise articles on their respective subjects. The chapter on Tuberculosis, though short, is commendable in its clarity. The section on Poliomyelitis is one of the book's best, presenting an interesting and brief discussion of physiotherapy based on the Sister Kenny Treatment. Dr. Top's obvious wealth of experience in Communicable Diseases binds the various chapters together and makes for good continuity. The photographs in colors and black and white from the Herman Kiefer Hospital in Detroit, and through the courtesy of the Parke, Davis and Company are excellent. The book reads easily and rapidly. Ample space is devoted to the details of nursing care.

There are still many instances wherein this volume is lacking. The chapter on Diphtheria does not mention the use of antihistaminics in the treatment of serum sickness following serum therapy. It neglects completely the use of the EKG in the early diagnosis and follow-up of diphtheritic myocarditis. It recommends the use of laryngeal intubation in preference to tracheotomy in the treatment of laryngeal diphtheria; the former procedure is considered by many others to be much more difficult to perform satisfactorily, and more fraught with dangers of complicating laryngeal stenosis. The section on Syphilis is perhaps justified in its consideration of penicillin therapy as experimental and without standardization at the time of its writing in January 1947. However, the continued recommendation that mercurial inunctions be used "in the accepted manner" in treating syphilis in late pregnancy and syphilis in infants is archaic. The appendix, an able compilation of interesting statistics on communicable diseases admitted to the Herman Kiefer Hospital during the years 1927 to 1936, would be far more valuable if comparative studies through the year 1946 were added.

Some of the chapters contain sufficient detail and depth to be of definite value to physician or medical student. Many portions are far too superficial. The

Glossary, containing definitions of such words as "adenitis", "bacteremia", "cachexia", and "debilitated", qualify the book, as does the preface to the first edition, as a text or "handy reference for all such persons whose professional duties necessitate contact with certain communicable diseases or infestations". It cannot be recommended as a student text or as a reference for physicians.

M. G.

Clinical Laboratory Methods and Diagnosis, Fourth Edition. By R. B. H. GRADWOHL. The C. V. Mosby Co., St. Louis Missouri, 1948. Illus. 4148 pp. $40.00.

This massive work in 3 volumes attempts to present descriptions of most of the laboratory tests and procedures available to the physician, as well as discussion of the clinical situation in which the procedures are employed. The extensive increase in the use of the laboratory in every branch of medicine has made it impossible for the clinical pathologist to keep abreast of developments in all fields. These volumes provide an example of that failure. In attempting to cover so much routine, individual subjects are not only superficially, but inaccurately treated. Tests are described without critical evaluation of their specificity or clinical value. The material for discussion on various diseases states is often poorly chosen. This text will not satisfy the physician or medical student. Its only value would appear to be as a reference book for the laboratory technician.

C. L. C.

Vol. III: This volume is worthy of separate review, because it contains so much new material and might well be considered as a separate text on Tropical Medicine or Clinical Parasitology. Dr. Pedro Kouri, who is Professor of Parasitology and Tropical Medicine at the Havana University Faculty of Medicine, Cuba has furnished a great number of excellent photographs. These are good and worthwhile and many of them are better than the usual ones presented in standard works on the subject.

There is a great need for a really satisfactory textbook on Tropical Medicine; one which might combine the basic parasitological knowledge of a biologist with that of a keen clinician, who has practiced for a number of years in tropical climes. This book does not fulfill these desiderata. Indeed there are a number of either great exaggerations of clinical pictures or gross errors. The description of the symptomatology of the "carrier" of amoeba cysts includes almost every possible neurasthenic symptom that man may have in the tropics. The statement that Endamoeba hystolytica has been recovered from duodenal washings is startling. The description of malaria is not only weak, but parts of it are contradictory and to the best of modern knowledge wrong. Sporozoites from the mosquito almost surely do not penetrate the red cells of man directly as indicated in the chart on pages 214 and 215. This pictorial representation which is based on Schaudinn's original description has not been confirmed, and there is a wealth of data showing that the parasite is not present in the blood from a short time after infection until

almost the end of the incubation period. In the discussion of enterobiasis on page 315 there is no indication of the great superiority of anal swabs of either cellophane or scotch tape when searching for eggs. One finds this in small print on page 53, contradicting the main text.

This, then, is a book which with a lot of work could be made into an excellent text.

F. B. B.

Viral and Rickettsial Infections of Man. Edited by THOMAS M. RIVERS, M.D., Director of the Hospital of the Rockefeller Institute for Medical Research. 77 illustrations, including 6 plates in color. J. B. Lippincott Company, Philadelphia-London-Montreal, publishers. 587 pages. $5.00.

Viral and Rickettsial Infections of Man, edited by T. M. Rivers, Director of the Hospital of the Rockefeller Institute for Medical Research and authored by a variety of experts at the Institute or elsewhere, is intended primarily for medical students and practicing physicians. As such, it fulfills very adequately a tremendous need for a simplified up-to-date textbook on these diseases. The chapters are well written and the information in each case is authoritative. Besides the chapters on a variety of different infections of man, there are included short chapters on physical and chemical procedures by Stanley and Lauffer; on serologic reactions by J. E. Smadel; chick embryo techniques by Goodpasture and Buddingh; propagation in tissue cultures by Enders, epidemiology by Maxcy and bacterial viruses by Hershey and Bronfenbrenner.

It is true that most of these particular chapters are of little practical use to the practicing physician in the diagnosis and treatment of the common virus diseases, but basic information is important in these diseases. Other chapters which cover specific diseases are of great practical and theoretic interest. Particularly welcome is the chapter on trachoma and inclusion conjunctivitis by Phillips Thygeson, for little of this information has gotten into the general literature. The chapter on dengue by Albert Sabin also is full of new and hitherto unpublished information. It is to be hoped that having written this chapter Dr. Sabin will be stimulated to publish the basic information on which it is founded. Otherwise workers in virus diseases will continue to have to accept the conclusions on faith.

In summary this is an excellent elementary text, which makes no attempt at covering all of the basic information on virus diseases in general, but is of real value in giving general concepts and summarizing knowledge.

F. B. B.

Bacterial and Mycotic Infections of Man. Edited by RENÉ J. DUBOS, PH.D., The Rockefeller Institute for Medical Research. 101 illustrations, including 3 plates in color. J. B. Lippincott Co., Philadelphia-London-Montreal, publishers. 785 pages. $5.00.

This is intended as a companion text to Viral and Rickettsial Infections of Man. It has been edited and written with much the same point of view. The need for

this book is not quite as great as that of the first, for there are a number of fairly adequate recent texts of bacteriology. However, most of the chapters in this book seem to be written with a slightly different viewpoint from the average textbook. The chapters on Parasitism and Disease by Francis, and Epidemiology by Maxcy give a clue to this, for particularly here is recognition given to the influence of Theobald Smith's concepts of the biological nature of disease. The individual chapters by individual authorities again present a high standard of excellence and range, from the general subjects to specific infections. Properties of Bacteria which Enable Them to Cause Disease by MacLeod and Pappenheimer, and the Allergic State by Merrill Chase deserve particular notice.

This book has not been written by clinicians and thus those who search in this text for up to date information on the treatment of some of the bacterial diseases will occasionally be disappointed. For instance, the statement on meningococcal meningitis "Penicillin may be as satisfactory as sulfadiazine; systemic administration of the drug should be supplemented by intrathecal route" (p. 514), does not adequately reflect the general use of combined sulfadiazine and penicillin for this infection. The table on p. 681 on the "Choice of Chemotherapeutic Agent" lists granuloma inguinale as if it were a virus disease and gives no indication of the established value of streptomycin in this bacterial disease. This is within a chapter which is excellent in general concepts and principles, but such minutiae are important when dealing with particular patients.

In summary this book, like its companion, is a real contribution and an excellent text, but should not be viewed as a clinical text, which it does not really pretend to be. The greatly reduced cost of these two texts is partly due to support from the National Foundation of Infantile Paralysis.

F. B. B.

Fundamentals of Psychiatry, Fourth Edition. 1948. By EDWARD A. STRECKER. J. B. Lippincott Company. 303 pages. $4.00.

The fourth edition of this standard compendium of psychiatry, revised and amplified, remains a very adequate presentation of descriptive psychiatry vitalized by simplified dynamic concepts. The simplifications of theory and case illustrations may be misleading. There is a strong tendency to cite possible organic causes of mental illness and to stress organic reaction patterns rather than enter into the complexities of personality development and its deviations. A brief new chapter on psychosomatic medicine deals with physiologic dysfunction due to emotional disorders rather than with medical illnesses in which emotional difficulties play an etiologic role. The book retains the easy but ineffectual means of disposing of various asocial and antisocial personalities by labeling them constitutional and lumping them with mental deficiency as defect states. Although the book has many merits as a brief review of well accepted concepts, it cannot be considered a reflection of current thinking concerning the dynamic approach to the understanding of personality function and its disorders.

T. L.

Handbook of Orthopaedic Surgery, 3rd edition. By ALFRED R. SHANDS, JR., in collaboration with RICHARD B. RANEY. Illus. 574 pp. $6.00. C. V. Mosby Co., St. Louis, Mo.

This book was very interesting to me. I have read small portions of the first and second editions from time to time and found them useful. The third edition, although it follows the pattern of the first two editions is more concise and complete. This is an excellent book for reference experience if one must prepare lectures to medical students or nurses. The bibliography is especially valuable in this regard. The numerous illustrations bring out the facts forcefully.

I heartily recommend the third edition to all who are interested in the diseases and disturbances of the musculo-skeletal system.

W. R. F.

Edinburgh Post-Graduate Lectures in Medicine. Volume IV, 1944–1947. Edinburgh, Oliver and Boyd. 582 pp. 18s. net.

This volume contains forty-one lectures on diverse unrelated subjects. The quality of the lectures varies tremendously. Much of the material can be better obtained from original sources or other texts. Some of the lectures suffer from a pontifical style which is likely to develop in presentations prepared for delivery.

The varied quality and interest in the articles should not detract from some which are excellent reviews and comments in their field. The volume is valuable only as a library collection of articles which have already been published separately.

E. N.

The Thyroid and Its Diseases, 2d ed. By J. H. MEANS. Illus. 571 pp. $12.00. J. B. Lippincott Co., Philadelphia, 1948.

The second edition of this popular text is a very welcome help in a field in which there has been tremendous activity and progress in the last ten years. The antithryoid drugs and radio-active iodine have been responsible for new therapeutic and investigative attack on the thyroid diseases. To write a text in the midst of such rapid developments requires nice judgement and selection of facts. The book's greatest value continues as—"an account of the personal experiences of a considerable group of workers . . . ". Every physician should have the book.

E. N.

Stethescopic Heart Records. Columbia Masterworks. Set M-735. $15.00.

These records played on a high fidelity phonograph give excellent results. The results are greatly dependent on the qualities of the amplifier and particularly the speaker in the phonograph machine used. A single large speaker will lose much of the high pitched sounds and accentuate or even rattle the low sounds. A small single speaker will better reproduce the higher pitched sounds (as well as the needle scratches and extraneous noises). Thus it is not easy to bring out all that is in these reproductions. They are particularly valuable for training in timing of

sounds, and with a good phonograph a student can learn much about the characteristics of the sounds. They can be studied and repeated often at leisure—the greatest advantage of records.

E. N.

Treatment of Heart Disease. By William A. Brams. Illus. 195 pp. $3.50. W. B. Saunders Company, Philadelphia, Pa. 1948.

In the preface Dr. Brams states that he intends this book to be "a systematic and practical guide in the treatment of heart disease" intended primarily for the general practitioner and the medical student. It succeeds admirably in this purpose for the practitioner but falls somewhat short for the medical student.

The author begins with a brief but sound discussion of the pharmacological action of drugs used in the treatment of heart disease, then discusses the treatment of congestive failure in general. Each major etiological group of heart disease is then discussed, again purely from the therapeutic standpoint, the author assuming in each case that the proper diagnosis has been made. He ends with the principles of management of cardiac patients during pregnancy and surgery.

The book is well organized, fairly comprehensive and very clearly written. Dr. Brams speaks with authority based on experience and familiarity with the literature. He admits that the opinions expressed are contested by some cardiologists, but actually there is very little argument to be found with the fundamental principles he expresses. Various cardiologists would make changes in his regimen of treatment of some diseases, but these differences would be based largely on individual preference for the use of certain drugs rather than on disbelief in Dr. Brams's regimen.

The index is well organized and adequate. The bibliography is organized numerically as the references are made in the text, hence serves no purpose as a source of information in itself.

For the practitioner who is interested in a brief, practical guide to treatment of cardiac disorders with a minimum of discussion of alternative regimens, this book will be found most useful. Likewise the medical student who is interested in a summary of commonly accepted and sound regimens for handling cardiac problems will find this book very helpful, but because of its lack of discussion of controversial points, its lack of presentation of different views on these points, and its meager reference to the literature, it should not be recommended to medical students as their sole text on the treatment of heart disease.

R. M.

Treatment by Diet, 5th edition. By CLIFFORD J. BARBORKA, M. D. Philadelphia: J. B. Lippincott Co. 1948. Cloth. Pp. 784. Price $10.00.

This book of 784 pages attests to the continued need for better and more widespread information on "Treatment by Diet". This new fifth adition shows the authors same conscientious attempt to simplify the subject as he demonstrated in preceding editions. It reveals an awareness, as the author states that "Intelli-

gent treatment by diet is the greatest weapon available to preventive medicine". In view of this declared concept one is surprised that the book is not as practical, as useful, as concise, as clear and as firmly founded on sound physiology and nutrition as the author undoubtedly intended it to be. In spots the text is ambiguous. For example on page 97 he states "When protamine zinc insulin is used alone it is often advisable to give about a 100 gram serving of 10 or 15% fruit at bed time". On page 98 he continues "When the morning administration of protamine zinc insulin is first commenced, it may sometimes be difficult to adjust the dose and the diet so as to avoid early morning hypoglycoemia. A light meal of a glass of milk or a cup of tea with soda biscuits shortly before bed time has been efficacious". Such statements are, to say the least, confusing. No where in the text is there mention of the desirability of the bed time meal being made up of food which will supply available glucose several hours after ingestion rather than immediately.

The author is to be commended upon keeping the protein in his high calorie diet lists well within normal limits. However he neglects to mention the importance of a high protein intake in cases of emaciation due to febrile disorders, etc. It seems lamentable that only one page is devoted to the subject of diet in pregnancy and lactation and that the advice given as to the modification of the normal diet in these two conditions is identical. It is obvious that the author has not correlated the Recommended Dietary Allowances of the National Research Council for pregnancy and lactation which he reprints on page 710 with the diet he recommends. His suggested addition of "one pint of milk" to the diet in health outlined on pages 58–59 fails to take care of the increased need of ascorbic acid in either condition or the demand for calcium and protein in lactation. The careful reader will observe that on page 604 the author discusses protein needs in pregnancy and lactation but he fails to mention on page 613 the advisability of referring to these comments so they will quite likely be lost to those using the book as a quick reference. One can not fail to wonder as to the selectivity employed in this book when less than one page is devoted to the discussion of diet in the omni-present conditions, pregnancy and lactation, while forty pages are given over to the less frequently used ketogenic diet.

In the treatment of pernicious anemia the author recommends liver in abundance "up to 500 grams a day". The efficacy of such therapy is unquestioned but that it has given way to more practical, less expensive and more acceptable therapy is widely recognized. There are ideas sprinkled throughout the book which seem out-moded. It is to be hoped that the sixth edition will be brought up to date which will enhance its usefulness.

E. R. T.

Principles Governing Eye Operating Room Procedures. By EMMA I. CLEVENGER. Illus. 215 pp. $5.50. The C. V. Mosby Co., St. Louis, Missouri.

Miss Clevenger's complete and careful presentation of operating room technique for nurses will be of great assistance as a guide to new departments in ophthal-

mology as well as a reference for established departments. The importance of the many small details of operating room management is emphasized as the basis of successful nursing support.

The excellent illustrations of the instrument tray set ups for the different procedures are especially valuable.

The manual will definitely take its place as an excellent reference book.

F. W.

BOOKS RECEIVED FOR REVIEW

Blood Clotting and Allied Problems. Transactions of the First Conference, Feb. 16–17, 1948. Publication of the *Josiah Macy, Jr. Foundation*, New York, N. Y. pp. 179.

Conditioned Reflexes and Neuron Organization. By Jerzy Konorski. Illus. 267 pp. $4.00, published Nov. 9, 1948 by *Cambridge University Press, The Macmillan Co.* New York, N. Y.

Diabetic Manual. By Elliott P. Joslin. Published by *Lea and Febiger*, 260 pp. illus. 1 plate in color. $2.50 (Oct. 1948)

Human Biochemistry. By Israel S. Kleiner. Published by *C. V. Mosby Co.* 1948, pp. 649. $7.00.

Intern's Manual. Published by *W. B. Saunders*, 201 pp. $2.25

Medical Statistics from Graunt to Farr. By Major Greenwood, *Cambridge University Press*, Nov. 9, 1948. $2.00

Pathology. By W. A. D. Anderson. Published by *C. V. Mosby Co.* 1948. 1453 pp. $15.00

Experimental Immunochemistry. By Elvin Kabat and Manifred Mayer, 567 pp. $8.75. Published by *Charles C. Thomas*, Springfield, Illinois.

Industrial Hygiene and Toxicology (in two volumes) Vol. 1, by Frank A. Patty, pp. 531. $10.00. Published by *Interscience Publishers, Inc.* 215 Fourth Ave. New York, N. Y.

Report of the Sanitary Commission of Massachusetts 1850. By Lemuel Shattuck and others. Published by the *Harvard University Press*, Cambridge, Massachusetts, 321 pp. $4.50

Biological Reactions Caused by Electric Currents and by X-rays. By J. Th. van der Werff, 203 pp. $5.00. Published by *Elsevier Publishing Co., Inc.* 215 Fourth Ave. New York 3, N. Y.

Cancer of the Esophagus and Gastric Cardia. By George T. Pack, published by *C. V. Mosby Co.* St. Louis, Missouri, 192 pp. $5.00

Factors Regulating Blood Pressure. Transactions of the Second Conference, Jan. 8–9, 1948, pp. 170. Publication of the *Josiah Macy, Jr. Foundation*, N. Y.

The Basis of Chemotherapy. By Thomas S. Work and Elizabeth Work, pp. 465. $6.50. Published by *Interscience Publishers Inc.*, New York

The Renal Origin of Hypertension. By Harry Goldblatt. Published by *Charles C. Thomas*, Springfield, Ill. 126 pp. $2.75

The Selected Writings of William Clowes. By F. N. L. Poynter, pp. 179, 15 s. Published by *Harvey and Blythe Lmtd.*, London, W. I.